Recent advances in brain tumor-targeted nano-drug delivery systems

Yu Liu & Weiyue Lu
†Fudan University, School of Pharmacy, Shanghai, China

Introduction: Brain tumors represent one of the most challenging and difficult areas in unmet medical needs. Fortunately, the past decade has seen momentous developments in brain tumor research in terms of brain tumor-targeted novel nano-drug delivery systems with significant important superiority over conventional formulations with respect to decreased toxicity and improved pharmacokinetics/pharmacodynamics.

Area covered: This review first introduces the characteristics of the two major obstacles in brain-tumor targeted delivery, blood–brain barrier (BBB) and blood–brain tumor barrier (BBTB), and then reviews recent advances in brain tumor-targeted novel nano-drug delivery systems according to their targeting strategies aimed at different stages of brain tumor development and growth.

Expert opinion: Based on continuously changing vascular characteristics of brain tumors at different development and growth stages, we propose the concept of ‘whole-process targeting’ for brain tumor for nano-drug delivery systems, referring to a series of overall targeted drug delivery strategies aimed at key points during the whole development of brain tumors.

Keywords: blood–brain barrier, blood–brain tumor barrier (BBTB), brain tumor, EPR effect, nano-drug delivery, whole-process targeting (WPT)


1. Introduction

Brain tumors greatly threaten human health for its fast development, poor prognosis and difficult treatment, causing a terrible mortality, which remains high through these years. To date chemotherapy has achieved limited success due to the low drug permeability from vessels into brain tumor tissue. In recent decades, brain tumor targeted nano-drug delivery systems attract more and more interest for their effectiveness in delivering drug to the exact foci and decreasing unnecessary drug accumulation in normal brain and peripheral tissue. This review will discuss recent advances in brain tumor-targeted nano-drug systems classified according to their targeting strategies aimed at different stages of brain tumor development and growth.

2. Brain tumors

According to the estimate of GLOBOCAN project 2008 (WHO), the global incidence of malignant brain cancer is about 3.5/100,000. The hazard of brain tumor to human health is rather terrible due to its poor prognosis, difficult control and wide age distribution [1]. For instance, glioblastoma multiforme [2], the most frequent primary central nervous system (CNS) tumor, represents the second cause of cancer death in adults less than 35 years of age [3]. Due to the invasive growth of brain tumor, the normal brain tissue will be suppressed, the CNS will be damaged and the life of the patient will be hazarded [4]. The survival time of brain tumor patients
3. Two obstacles to brain tumor treatment: blood–brain barrier and blood–brain tumor barrier

The effectiveness of systemic chemotherapy on brain cancer is usually very limited due to the hindrance to drug permeability from vessels into tumor tissues caused by the two barriers unique to the brain: blood–brain barrier (BBB) and blood–brain tumor barrier (BBTB).

BBB is made up by the capillary endothelial cells that line the cerebral microvessels and surrounding perivascular elements such as basal lamina, pericyte, astrocyte end feet and interneurons and separates the brain from the rest of the body with tight junctions [15]. The transendothelial electrical resistance (TEER) of the BBB is estimated to be 8,000 Ω/cm² [16], precluding the access for almost all large molecule drugs and more than 98% small-molecule candidate drugs to the brain tissue [17]. To deliver therapeutic agents across the BBB, mechanisms for transport of endogenous molecules into the brain have been employed such as receptor-mediated endocytosis, adsorptive-mediated transcytosis, transporter uptake and membrane permeation of lipophilic molecules. The receptor-mediated endocytosis represents the most widely used strategy for brain-targeted drug delivery [18]. Various receptors and transporters are present on the luminal endothelial plasma membranes, including the transferrin receptor [19], the insulin receptor [20], endothelial growth factor receptor [21], low-density lipoprotein receptor [22] and glucose transporter [23] providing binding targets for drug delivery systems.

The permeability of brain tumor vasculature changes with the whole development progress of tumors [24]. At the early stage of malignant brain tumor such as glioma or small brain metastasis, the growth of tumor cells depends on normal brain vascular systems before the formation of tumor neovessels and the BBB remains intact. With the deterioration of brain tumor, tumor cells begin to invade the surrounding normal brain tissues. Only when the tumor cell cluster reached a volume large enough (> 0.2 mm³) will BBB be damaged and BBTB be formed. BBTB exists between the brain tumor tissues and capillary vessels, preventing the delivery of most hydrophilic molecules and antitumor agents to brain tumor [25]. Brain tumor neovasculature is functionally different from both normal brain capillaries and the neovasculature of peripheral tumors. In comparison with normal brain capillaries, the capillary vessels of brain tumors include continuous fenestrated microvessels [26] with the critical pore for solute permeability being about 12 nm as demonstrated by studies with dendrimers with different molecular sizes. Only spherical nanoparticles smaller than 12 nm can pass fenestrated microvessels constituting BBTB [27].

With the further development progress of brain tumor, when both BBB and BBTB become impaired, endothelial gaps form on the microvessels of brain tumors and enhanced permeability and retention (EPR) effect comes into action, resulting in the tendency of nanoscale particles (typically liposomes [28], nanoparticles [29] and macromolecular drugs [30,31]) to accumulate in tumor tissue much more than they do in normal tissues. However, the blood vessel permeability of intracranial solid tumor is far lower than that of peripheral solid tumor perhaps because of the relative scarce presence of trans-endothelial cell gaps, caveolae and organelles related with vesicles. In other words, the critical pore size for
solute passage of intracranial tumors is almost always smaller than peripheral tumors. For example, the critical pore size for intracranial tumor model of murine hepatocarcinoma (HCA-1), mouse mammary carcinoma (Shionogi) and mouse MCA-IV breast carcinoma cell lines were reported to be 210–380, 100–380 and 380–550 nm, respectively, much smaller than those for peripheral model (380–550, 200–380, 1200–2000, respectively). And the critical pore size for intracranial human primary glioblastoma cell line U87MG is as low as 7–100 nm [32]. All these differences of critical pore size for solute passage suggest the relative weak EPR effect of brain tumors.

Good treatment of brain tumor-targeted drug delivery should be able to localize the therapeutic agents at the brain tumor foci and at the same time minimize the involvement of peripheral tissues and healthy brain tissue [33,34]. That is why nanovehicles received so much attention these years for their controllable particle size and easy surface modification. This review will discuss recent advances in brain tumor-targeted nano-drug systems classified according to their targeting strategies aimed at different stages of brain tumor development and growth: i) BBB targeting, that is, trans-BBB drug delivery and further tumor foci-targeting drug delivery, for early stage of brain tumor when the BBB is still intact; ii) BBTB targeting for brain tumors with angiogenesis and iii) particle size controlling based on EPR effect for brain tumors with impaired vascular endothelium.

4. Brain tumor-targeting strategies and related nano-drug delivery systems

4.1 BBB-targeting strategies and related nano-drug delivery systems

4.1.1 Trans-BBB drug delivery for brain tumor treatment

4.1.1.1 Receptor-mediated trans-BBB drug delivery

Many types of receptors existing on the BBB can be used as the target of ligand-mediated trans-BBB transport. For example, nicotine acetylcholine receptors (nAChRs) are ligand-gated ion channels expressed mainly in the nervous system and at the neuromuscular junction. It is widely expressed in the brain, including the brain capillary endothelial cells (BCECs). Therefore, nAChRs mediate brain transport may be a promising strategy. Neurotoxins from snake venoms are well known to bind with high affinity and selectivity to nicotinic acetylcholine receptors. They belong to a family of proteins called ‘three-finger toxins,’ which adopt a flat, leaf-like shape formed by three adjacent loops that emerge from a small globular core. The tips of the loop II of neurotoxins are thought to be nAChR-binding domains. Based on the above facts, our group conducted a series of investigation on novel polypeptide-mediated brain tumor-targeted nano-drug delivery systems [35,36]. Firstly, brain-targeting functional polypeptides were screened with computer-aided drug design (CADD) using loop II sequence and loop II tip sequence as the lead. Considering the similarity of acetylcholine-binding protein (AChBP) with central nicotine acetylcholine receptor (nAChR α7), AChBP was used as the model receptor molecules. Three brain-targeting amino acid sequences RVG29 (29 amino acid residues), KC20 (20 amino acid residues) and CDX (16 amino acid residues) were designed and optimized. In the in vitro cellular uptake experiment, all the three functional polypeptides labeled with fluorescein isothiocyanate (FITC) could be
selectively uptaken by BCEC cells, different from the negative control group nAChR-absent HeLa cells. The binding affinity with nAChRs for three polypeptides was in the following order: KC20.25 > RVG29 > CDX. In the in vivo imaging monitoring, FITC-labeled CDX, KC20.25 and RVG29 all concentrated in the brain and kidney, exhibiting considerable brain-target ability. Among the three peptides, CDX was chosen for modification of micellar materials poly(ethylene glycol)-poly(lactide) (PEG-PLA) due to its good tissue selectivity and the shortest sequence. CDX modification significantly enhanced the fluorescent strength in the brain after intravenous injection in mice of 6-coumarin-containing micelles with the maximal drug concentration (Cmax) and area under curve (AUC) in the brain being 2.1 and 1.9 times those for unmodified micelles (p < 0.05). In glioma-bearing nude mice, the glioma-targeting efficiency by CDX modification was proven by the following in vivo fluorescence imaging. CDX-modified micelles entered the brain 2 h after injection and its brain distribution lasted more than 4 days. Finally, let us return to the gold standard: pharmacodynamic evaluation. After loading with paclitaxel (PTX), CDX-modified PEG-PLA micelles showed the best survival outcome with the medium survival time of 27 days after intravenous injection to nude mice bearing intracranial glioma U87MG on 5, 10, 15 days after tumor implantation, significantly longer than unmodified PTX-loaded PEG-PLA micelle (20 days) and normal saline (18 days).

Since the first introduction of the in vivo phage display by Pasqualini in 1996 [37], this technique has been expanded and has provided tissue-specific peptides as targeting moieties for tumors and organs [38-40]. In searching for novel targeting peptide, in vivo phage display has also been proven high throughput and efficiency [41]. For example, Li et al. employed a 12-mer phage display peptide library to isolate a peptide (denoted as Pep TGN) for brain targeting. Pep TGN was covalently conjugated onto the surface of poly(ethylene glycol)-poly(lactic-co-glycolic acid) (PEG-PLGA). The in vitro bEnd.3 cellular uptake of Pep TGN decorated nanoparticles was significantly higher than that of unmodified nanoparticles. Enhanced brain accumulation efficiency together with lower accumulation in liver and spleen was observed in the nude mice intravenously injected with Pep TGN-conjugated nanoparticles, showing powerful brain selectivity of Pep TGN. The brain Drug Targeting Index (DTI) of coumarin incorporated in targeted nanoparticles was significantly higher than that of coumarin incorporated in plain nanoparticles [42].

4.1.1.2 Adsorptive-mediated trans-BBB drug delivery

Cationic bovine serum albumin (CBSA)-mediated trans-BBB transport of nanoparticles is a typical example of absorptive-mediated transcytosis. Lu et al. first reported a CBSA-modified PEG-PLA nanoparticle (CBSA-NP) for brain delivery. They found that increasing the surface CBSA density of the nanoparticle enhanced the BBB permeability-surface area but decreased blood AUC and accelerated blood clearance [43]. After loading plasmid pORF-hTRAIL (pDNA) for gene therapy of gliomas, CBSA-modified hTRAIL-loaded nanoparticles achieved good transfection effect both in vitro and in vivo. In vivo it was first internalized by C6 glioma cells and located in the cytoplasm 30 min after transfection, in contrast to unmodified NP-hTRAIL, which was entrapped in the endolysosomal compartment. At 6 and 48 h after transfection, released pDNA was found in the nuclei and induced apoptosis. At 30 min after i.v. administration of CBSA-modified hTRAIL-loaded nanoparticle to BALB/c mice bearing i.c. C6 gliomas, CBSA-modified hTRAIL-loaded nanoparticle was colocalized with glycoproteins in brain and tumor microvasculature and via absorptive-mediated transcytosis, accumulated in tumor cells. At 24 and 48 h after i.v. administration of CBSA-modified hTRAIL-loaded nanoparticle, respectively, hTRAIL mRNA and protein were detected in normal brain and tumors. Furthermore, repeated i.v. injections of CBSA-modified hTRAIL-loaded nanoparticle induced apoptosis in vivo and significantly delayed tumor growth [44]. Later Agarwal et al. used solid lipid nanoparticles (SLNs) for CBSA modification to bypass BBB and provided improved therapeutic efficacy of encapsulated anticancer drug methotrexate (MTX). Transendothelial transport study on brain capillary endothelial cells (BCECs) depicted CBSA-conjugated SLNs to undergo transcytosis to a greatest extent. These SLNs were preferably taken up by BCs and human neuroglial culture (HNGC)-1 tumor cells as evaluated against unconjugated SLNs and plain MTX. Furthermore, cytotoxicity studies were performed on HNGC1 tumor cells. CBSA-conjugated SLNs exhibited more potent cytotoxic effect on HNGC1 cells than free MTX [45].

Polysorbate 80 coating of nanovesicles can also act with BBB and result in trans-BBB transport. Poly(n-butylcyanoacrylate) nanoparticles (PBCA-NP) has been used for a long time for brain-targeting drug delivery systems and polysorbate 80 coating has been proven to help nanoparticles to cross BBB. The mechanism of the influence of polysorbate 80 on BBB has been discussed in a recent study, which monitored the development of TEER after the addition of PBCA-NP employing impedance spectroscopy and verified the integrity of the BBB in vitro by measuring the passage of the reference substances 14C-sucrose and FITC-BSA (bovine serum albumin) after addition of PBCA-NP. Application of polysorbate 80-coated PBCA-NP leads to a reversible disruption of BBB after an interaction of 4 h [46]. In addition, the use of polysorbate 80 coating is not limited to PBCA nanoparticles. Soni et al. used a random copolymeric micelles composed of N-isopropylacrylamide and N-vinylpyrrolidone cross-linked with N,N′-methylenebisacrylamide as nanogel carriers to encapsulate N-hexylcarbamoyl-5-fluorouracil, a prodrug of fluorouracil, which could be targeted to brain tissue across BBB after coating with polysorbate 80. Gamma Scintigraphy in mice and rabbits found that polysorbate 80 coating could change the surface characteristics of
nanogels, increasing intracranial accumulation from 0.18% for unmodified nanogels to 0.52% for modified nanogels [47].

4.1.1.3 Lipophilicity-based trans-BBB transport
Myristic acid (MC) is a fatty acid with high hydrophobicity sufficient to be incorporated into the fatty acyl core in the phospholipid bilayer of the eukaryotic plasma membrane of the cell and can act as a lipid anchor to biomembranes. It can increase the brain uptake, as demonstrated by the successful transport of myristoylated polyarginine crossing the BBB and the resulted significant accumulation in neuron [48]. The brain tumor-targeting capability of MC has been proven by Li et al., who modified polyethylene glycol (PEG) with MC to increase the brain uptake, transfection and gene expression using polyethyleneimine (PEI) as the gene carrier in vitro and in vivo. The anti-glioblastoma effect of pORF-hTRAIL myristic acid-modified polyethyleneimine complex (MC-PEI/pORF-hTRAIL) was demonstrated by the prolonged mean survival time of intracranial U87MG glioblastoma-bearing mice (MC-PEI/pORF-hTRAIL 28 days versus unmodified pORF-hTRAIL-polyethyleneimine complex 24 days, the myristic acid-modified polyethyleneimine-control reporter gene (pGL) complex group 21 days and normal saline 22 days) [49].

4.1.1.4 Transporter-mediated trans-BBB drug delivery
Glucose transporter (GLUT-1) is found in high density on BBB [50] and various tumors [51], conferring brain tumor-targeting property through facilitative glucose metabolism by the glucose transporters [52]. So the attachment of glucose molecules to dendrimers enabled its recognition by GLUT1 transporters. For instance, Dhanikula et al. evaluated polyether-copolyester (PEPE) dendrimers as methotrexate carriers for the treatment of gliomas and conjugated it to D-glucosamine as the ligand aimed at GLUT-1 for enhancing BBB permeability and tumor targeting. The efficacy of such MTX-loaded dendrimers was established against U87MG and U343 MGa cells. Glucosylated PEPE dendrimers were found to be endocytosed in significantly higher amounts than nonglucosylated PEPE dendrimers by both U87MG and U343 MGa cells and showed lower IC_{50} after MTX loading, suggesting that loading MTX in glucosylated PEPE dendrimers increased its potency. These MTX-loaded glucosylated dendrimers were able to kill even MTX-resistant cells highlighting their ability to overcome MTX resistance. In addition, the amount of MTX transported across the in vitro BBB model was increased by three to five times more after loading in the dendrimers and further increased by glucosylation. Glucosylated PEPE dendrimers distributed faster through out the avascular tumor spheroids than nonglucosylated PEPE dendrimers [53].

4.1.1.5 Cell-penetrating peptide-mediated trans-BBB transport
Cell-penetrating peptides (CPPs) are short amphipathic and cationic peptides that are rapidly internalized across cell membranes. They can be used to deliver molecular cargo, such as imaging agents (fluorescent dyes and quantum dots), drugs, liposomes, peptide/protein, oligonucleotide/DNA/RNA, nanoparticles and bacteriophage into cells [54]. TAT, the transactivating protein of the human immunodeficiency virus type-1 essential for viral replication, is perhaps the most frequently used CPPs for drug delivery system modification for its cationic charges facilitate the interaction with the normally negatively charged BBB. It can trigger permeabilization of the cell membrane via a receptor/transporter-independent pathway, which results in endocytosis of the sequence [55]. A TAT-modified liposomal formulation was prepared from TAT-conjugated cholesterol and showed strong inhibitory effect against C6s cell lines, higher efficiency of brain delivery, longer survival time of brain glioma-bearing animals and lower cardiotoxic risk [56]. To improve the brain delivery capability of dendrimer, polyamidoamine (PAMAM) dendrimer and TAT peptides were conjugated to bacterial magnetic nanoparticles (BMPs) for the construction of an efficient and brain-targeted gene delivery system (TAT-BMPs-PAMAM). Small interfering RNA expression plasmid (psiRNA) corresponding to the open reading frame of the human epidermal growth factor receptor gene (psiRNA-EGFR) to downregulate the EGFR gene was selected as the model drug. Compared with control groups, TAT-BMPs-PAMAM complex with psiRNA-EGFR resulted in better suppression of EGFR expression and a more obviously arrested effect on the proliferation and invasion ability of U251 cells in vitro and significantly suppressed growth rate of tumor and obviously downregulated expression of oncoproteins (EGFR, pAKT, MMP2/9, PCNA, VEGF, Bcl-2 and cyclin D1) in the U251 subcutaneous nude mouse model in vivo [57]. What should be pointed out for CPP modification is that CPP modification tends to enhance penetration into cells without cell specificity, resulting in simultaneous increase in drug concentration in both brain tumors and healthy tissues.

4.1.2 Trans-BBB plus brain tumor cell dual-targeting drug delivery
For better brain tumor delivery, researchers have combined trans-BBB targeting and brain tumor cell targeting by the following two principles: i) dual-targeting moieties modification, such as wheat germ agglutinin (WGA) plus transferrin (TF), mannoylpranoside plus TF and so on; ii) modification with a single-targeting moiety that targets both BBB and tumor cells, such as angioprep-2, TF and lactoferin (Lf) and so forth.

Let us first see several examples of dual-targeting modification. WGA seems to be a good candidate for drug carrier targeting BBB for low cytotoxicity and high affinity for the cerebral capillary endothelium compared with other lectins [58]. WGA has been reported to be co-modified with TF on the periphery of pegylated polyamidoamine (PAMAM-PEG) to get the PAMAM-PEG-WGA-TF conjugate (PAMAM-PEG-WGA-TF) to load DOX in the interior. TR is a cell membrane-associated glycoprotein involved in the cellular
uptake of iron and in the regulation of cell growth, and its expression in tumor and brain capillary endothelial cells is often elevated, making Tf and anti-Tf receptor antibody an excellent option for trans-BBB as well as brain tumor cell-targeting modification [59]. PAMAM-PEG-WGA-Tf delivered more DOX (13.5%) across in vitro BBB model than WGA-modified PAMAM-PEG (8%), for Tf-modified PAMAM-PEG (7%) and free DOX (5%) in 2 h and efficiently inhibited growth of glioma cell lines [60]. Another example is co-modifying topotecan liposomes with WGA plus taxotrex (TAM), a multidrug resistance reversing agent, which were reported to inhibit the drug efflux transporters expressed on the brain endothelial cells and on tumor cells [61], as reported by Du et al. The dual-modified liposome exhibited a significant inhibitory effect on glioma cells and capability of crossing BBB compared with unmodified topotecan liposomes. In the brain tumor-bearing rats, the dual-targeting effects in vivo of topotecan liposomes modified with TAM and WGA could be evidently observed, resulting in a significant improvement in the overall survival of the brain tumor-bearing rats compared with free topotecan and topotecan liposomes. Moreover, results from an extended treatment group indicated that the survival could be further significantly enhanced, indicating that an extended chemotherapy with topotecan liposomes modified with TAM and WGA would be beneficial for treatment. The dual-targeting effects in vivo of topotecan liposomes modified with TAM and WGA could be related to an enhanced effect by TAM via inhibiting efflux of MDR proteins in the BBB and the brain tumor, and an enhanced effect by WGA via endocytosis in the BBB and in the brain tumor [62]. TAM could be also co-modified on epirubicin liposomes with transferrin to obtain efficient inhibition on glioma cells or glioma spheroids in vitro, significant transport ability across the BBB model in vitro, an evident effect of targeting the brain tumor cells in vivo and an extended median survival time in the brain glioma-bearing rats [63].

Tf and p-aminophenyl-ε-D-manno-pyranoside (MAN) dual-modified daunorubicin liposomes have been designed by Ying et al. to make use of GULT-1 on BBB and TfR on glioma cells for brain tumor targeting (Table 1). The dual-targeting modification increased the transport ratio of daunorubicin liposomes across the BBB model up to 24.9% on the in vitro BBB model, enhanced the inhibitory rate to C6 glioma cells after crossing the BBB up to 64.0% and significantly prolonged the median survival time of tumor-bearing rats (22 vs 17 days for free daunorubicin, 18 days for unmodified liposomes, 19 days for MAN-modified liposomes and 18 days for Tf-modified liposomes) [63].

Angiopep-2 has been proven to be a dual-functional brain tumor-targeting ligand aimed at the low-density lipoprotein receptor-related protein (LRP), which is overexpressed on both BBB and glioma cells [64,65]. For example, we prepared an angiopep-2-modified PEG-PLA micelles with high brain accumulation in 24 h [66]. Our colleagues Xin et al. reported an angiopep-2-modified poly (ethylene glycol)-co-poly (epsilon-caprolactone) (PEG-PCL) nanoparticle drug delivery system (ANG-NP). Compared with nontargeting nanoparticles, a significantly higher amount of rhodamine isothiocyanate-labeled dual-targeting nanoparticles were endocytosed by U87MG cells. The antiproliferative and cell apoptosis assay of PTX-loaded ANG-NP (ANG-NP-PTX) demonstrated that ANG-NP-PTX resulted in enhanced inhibitory effects to U87MG glioma cells. In addition, the transport ratios across the in vitro BBB model were significantly increased by ANG-NP-PTX. Enhanced accumulation of ANG-NP in the glioma bed and infiltrating margin of intracranial U87MG glioma tumor-bearing in vivo model were observed by real-time fluorescence image [67]. To further realize the selective targeting to brain neuroglial cells in order to enhance therapeutic effects and limit side effects in nontarget cells, Gao et al. modified PEG-PCL nanoparticles (AENP) with both angiopep-2 and EGFP-EGF1 (a fusion protein derived from coagulation factor δ, which retains the specific Tf-binding capacity but does not cause coagulation) [68]. Angiopep-2 penetrates BBB and EGFP-EGF1 binds neuroglial cells. Both b.End.3 cells and neuroglial cells uptake more dual-targeted nanoparticles (AENP) compared with unmodified nanoparticles. ex vivo imaging showed that AENP had higher accumulation in the brain over unmodified nanoparticles and EGFP-EGF1 modified nanoparticles. Fluorescent in situ hybridization of brain slides demonstrated that AENP co-localized with neuroglial cells. Transmission electron microscopy further showed that AENP could target and enter neuroglial cells [69]. DOX-loaded angiopep-2-modified liposomes have also been proven stable and suitable for brain-targeting delivery by the smallest average tumor volume, the strongest tumor apoptosis, more weight gain after treatment and longer median survival time in glial mouse model [70].

Chlorotoxin (CTx), a 36-amino acid peptide originally isolated from Leirus quinquestriatus scorpion venom and highly toxic to invertebrates but non-toxic to mammals, is another brain tumor-specific targeting moiety for nano-drug delivery systems. It exhibited high specificity, selectivity and affinity for gliomas [71] and other tumors of neuroectodermal origin [72] through binding to a lipid raft-anchored complex that mainly contained the glioma-specific chloride ion channel and matrix metalloproteinase 2 (MMP-2) endopeptidase [73]. Xiang et al. employed CTx as the targeting ligand in preparation of brain glioma-targeted doxorubicin-loaded liposomes. CTxX highly facilitated the uptake of liposomes by and increased the cytotoxicity against three glioma cell lines (C6, U87MG, U251) and a murine microvascular endothelium cell line (BMECs). In BALB/c mice bearing U87 tumor xenografts, CTx-modified liposomes showed more accumulation in the subcutaneous and intracranial tumors, higher tumor growth inhibition and lower blood toxicity in the armpit tumor model [74].

Using TfR as the targeting goal and polymersomes [75] with higher stability compared with liposomes as the drug carrier, Pang et al. prepared a glioma-targeted Tf-conjugated biodegradable PEG-PCL polymersomes (TF-PO) with doxorubicin (DOX) as the model drug. Compared with unmodified DOX
Table 1. Targeting moieties on nano-drug carriers for brain tumor targeting.

<table>
<thead>
<tr>
<th>Targeting moeity</th>
<th>Carrier type</th>
<th>Polymers</th>
<th>Model drug</th>
<th>Outcome</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>BBB-targeting strategies and related nano-drug delivery systems</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Receptor-mediated trans-BBB drug delivery</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CDX</td>
<td>Micelles</td>
<td>PEG-PLA</td>
<td>Paclitaxel</td>
<td>Enhanced survival time of U87-bearing mice</td>
<td>[35,36]</td>
</tr>
<tr>
<td>Pep TGN</td>
<td>Nanoparticles</td>
<td>PEG-PLGA</td>
<td>Fluorescent probes</td>
<td>Enhanced brain accumulation, lower accumulation in liver and spleen</td>
<td>[42]</td>
</tr>
<tr>
<td><strong>Adsorptive-mediated trans-BBB drug delivery</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cationic bovine serum albumin</td>
<td>Nanoparticle</td>
<td>PEG-PLA</td>
<td>Plasmid pORF-hTRAIL</td>
<td>Induction of apoptosis \textit{in vivo} and significantly delayed tumor growth in BALB/c mice bearing i.c. C6 gliomas</td>
<td>[43,44]</td>
</tr>
<tr>
<td>Solid lipid nanoparticles</td>
<td>Lipids</td>
<td>methotrexate</td>
<td></td>
<td>Preferred uptake by BCs and human neuroglial culture (HNGC)-1 tumor cells, higher cytotoxicity on HNGC1 tumor cells</td>
<td>[45]</td>
</tr>
<tr>
<td>Polysorbate 80</td>
<td>Nanoparticles</td>
<td>N-isopropylacrylamide and N-vinylpyrrolidone</td>
<td>N-hexylcarbamoyl-5-fluorouracil</td>
<td>Increased intracranial accumulation</td>
<td>[47]</td>
</tr>
<tr>
<td><strong>Lipophilicity-based trans-BBB transport</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Myristic acid</td>
<td>PEI-gene complex</td>
<td>PEG-PEI</td>
<td>pORF-hTRAIL gene</td>
<td>Increased brain uptake, transfection and gene expression</td>
<td>[49]</td>
</tr>
<tr>
<td><strong>Transporter-mediated trans-BBB drug delivery</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>o-glucosamine</td>
<td>Dendrimer complex</td>
<td>Polyether-copolyester (PEPE) dendrimers</td>
<td>Methotrexate</td>
<td>Higher \textit{in vitro} cytotoxic and trans-BBB permeability and faster distribution throughout the avascular tumor spheroids</td>
<td>[53]</td>
</tr>
<tr>
<td><strong>Cell-penetrating peptides (CPPs)-mediated trans-BBB transport</strong></td>
<td>Liposome</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TAT</td>
<td>PAMAM-bacterial magnetic nanoparticles</td>
<td>PAMAM</td>
<td>psiRNA-EGFR</td>
<td>Strong inhibitory effect against C6s cell lines, higher efficiency of brain delivery, longer survival time of brain glioma-bearing animals and lower cardiotoxic risk</td>
<td>[56]</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Better suppression of EGFR expression, obviously arrested effect on the proliferation and invasion ability of U251 cells \textit{in vitro}, significantly suppressed growth rate of tumor and obviously downregulated expression of oncoproteins in the U251 subcutaneous nude mouse model \textit{in vivo}</td>
<td>[57]</td>
</tr>
<tr>
<td>Targeting moiety</td>
<td>Carrier type</td>
<td>Polymers</td>
<td>Model drug</td>
<td>Outcome</td>
<td>Ref.</td>
</tr>
<tr>
<td>------------------</td>
<td>-------------</td>
<td>----------</td>
<td>------------</td>
<td>---------</td>
<td>------</td>
</tr>
<tr>
<td>Trans-BBB plus brain tumor cell dual-targeting drug delivery</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>WGA + Tf</td>
<td>Dendrimer complex</td>
<td>PAMAM-PEG</td>
<td>Doxorubicin</td>
<td>More trans-BBB transport <em>in vitro</em></td>
<td>[60]</td>
</tr>
<tr>
<td>WGA + tamoxifen</td>
<td>Liposome</td>
<td>Topotecan</td>
<td></td>
<td>Significantly improved overall survival of the brain tumor-bearing rats</td>
<td>[62]</td>
</tr>
<tr>
<td>Tamoxifen + transferrin</td>
<td>Liposome</td>
<td>Epirubicin</td>
<td></td>
<td>Efficient inhibition on glioma cells or glioma spheroids <em>in vitro</em>, significant transport ability across the BBB model <em>in vitro</em> and an extended median survival time in the brain glioma-bearing rats</td>
<td>[63]</td>
</tr>
<tr>
<td>Tf + p-aminophenyl-α-L-manno-pyranoside</td>
<td>Liposome</td>
<td>Daunorubicin</td>
<td></td>
<td>Increased transport ratio of daunorubicin liposomes across the <em>in vitro</em> BBB model, enhanced inhibitory rate to C6 glioma cells after crossing the BBB and significantly prolonged the median survival time of tumor-bearing rats</td>
<td>[63]</td>
</tr>
<tr>
<td>Angiopep-2</td>
<td>Micelles</td>
<td>PEG-PLA</td>
<td>Fluorescent probes</td>
<td>High brain accumulation in 24 h</td>
<td>[66]</td>
</tr>
<tr>
<td>Angiopep-2 peptide + EGFP-EGF1</td>
<td>Nanoparticle</td>
<td>PEG-PCL</td>
<td>Paclitaxel</td>
<td>Enhanced inhibitory effects on U87MG glioma cells <em>in vitro</em>, enhanced accumulation in the glioma bed and infiltrating margin of intracranial U87MG glioma tumor-bearing <em>in vivo</em> model</td>
<td>[67]</td>
</tr>
<tr>
<td>Chlorotoxin</td>
<td>Liposome</td>
<td>Doxorubicin</td>
<td></td>
<td>The smallest average tumor volume, the strongest tumor apoptosis, more weight gain after treatment and longer median survival time in glioma mouse model</td>
<td>[70]</td>
</tr>
<tr>
<td>Tf</td>
<td>Polymersome</td>
<td>PEG-PCL</td>
<td>Doxorubicin</td>
<td>Strong cytotoxicity against C6 glioma cells <em>in vivo</em>, increased <em>in vitro</em> cytotoxicity against three glioma cell lines, accumulation in the subcutaneous and intracranial tumors, higher tumor growth inhibition and lower blood toxicity in the armpit tumor model</td>
<td>[74]</td>
</tr>
<tr>
<td>Anti-TfR antibody</td>
<td>Liposome</td>
<td>LaZ gene</td>
<td></td>
<td>Brain-specific anti-TfR antibody OX26-pGFAP modification resulted in less peripheral distribution and comparative brain tumor distribution than OX-26</td>
<td>[78]</td>
</tr>
<tr>
<td>Lactoferrin</td>
<td>Polymersome</td>
<td>PEG-PCL</td>
<td>Doxorubicin</td>
<td>Inhibited tumor growth and prolonged median survival time in glioma-bearing model mice</td>
<td>[81]</td>
</tr>
</tbody>
</table>
Table 1. Targeting moieties on nano-drug carriers for brain tumor targeting (continued).

<table>
<thead>
<tr>
<th>Targeting moiety</th>
<th>Carrier type</th>
<th>Polymers</th>
<th>Model drug</th>
<th>Outcome</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>BBTB-targeting strategies and related nano-drug delivery systems</td>
<td>Angiogenesis-specific targeted drug delivery</td>
<td>RGD</td>
<td>Micelle</td>
<td>PEG-PLA</td>
<td>Paclitaxel</td>
</tr>
<tr>
<td></td>
<td>Covalent conjugate</td>
<td>PAMAM</td>
<td>Doxorubicin</td>
<td>Accumulation preference at the brain tumor over nontumor tissue and prolonged the survival time of experimental glioma-bearing mice</td>
<td>[89]</td>
</tr>
<tr>
<td></td>
<td>Hollow gold nanosphere</td>
<td></td>
<td></td>
<td>A 20.7°C elevation in tumor temperature and a prolonged survival of tumor-bearing mice</td>
<td>[91]</td>
</tr>
<tr>
<td></td>
<td>DNA aptamer</td>
<td>Nanoparticle</td>
<td>PEG-PLGA</td>
<td>Enhanced cellular association of nanoparticles in C6 glioma cells, prolonged circulation and enhanced PTX accumulation at the tumor site</td>
<td>[94]</td>
</tr>
<tr>
<td>EPR effect-based strategies and related nano-drug delivery systems</td>
<td>Anti-EGFR antibody</td>
<td>Liposome</td>
<td>Sodium borocaptate</td>
<td>Distribution of the liposomes and the BSH in the tumor</td>
<td>[103]</td>
</tr>
<tr>
<td></td>
<td>Cationic solid lipid nanoparticle</td>
<td>Cacao butter and stearic acid</td>
<td>Carmustine</td>
<td>Effective <em>in vitro</em> delivery to U87MG cells and <em>in vivo</em> antiproliferative efficacy against the growth of malignant brain tumors</td>
<td>[104]</td>
</tr>
<tr>
<td></td>
<td>Epidermal growth factor</td>
<td>Au nanoparticle</td>
<td></td>
<td>10-fold improved selectivity to the brain tumor compared with untargeted conjugates</td>
<td>[105]</td>
</tr>
</tbody>
</table>
polysomes and free DOX, Tf-PO-DOX demonstrated the strongest cytotoxicity against C6 glioma cells in vitro, significantly reduced tumor volume, increased median survival time and extensive tumor cell apoptosis in vivo. Tf-PO also significantly enhanced brain delivery of DOX, especially the delivery of DOX into brain tumor cells [76]. Further research about the polysome uptake mechanism revealed that Tf-modified PO-DOX was mainly uptaken through a clathrin-mediated energy-dependent endocytosis and then intracellularly transported involving both the Golgi apparatus and lysosomes [77]. Besides transferrin, anti-TIR antibody OX26 has been widely used in preparation of immunoliposomes. However, the brain tumor selectivity of such OX26-modified immunoliposomes could not be assured, as shown by the report by Zhao et al., because parenchyma cells in the liver and spleen also express transferrin. So an anti-TIR specific for the brain provided a better choice for modification. Liposomes decorated with a brain-specific anti-transferrin antibody OX26-pGFAP (glial fibrillary acidic protein promoter) was able to obtain less peripheral 545 distribution and comparative brain tumor distribution in comparison with OX26-liposomes [78].

Lactoferrin (Lf) is a novel brain-targeting ligand that has been reported to transport across the BBB with high efficiency superior to transferrin [79]. More importantly, the receptor of Lf, low-density lipoprotein receptor-related protein (LRP), has been proven to be also overexpressed in glioma cells [80]. Therefore, a lactoferrin-conjugated PEG-PCL polymersome drug delivery system holding doxorubicin and MDR inhibitor tetrandrine (Lf-PO-Dox/Tet) demonstrated strong cytotoxicity against C6 glioma cells. Lf-modified polymersome could enter the brain and accumulate at the tumor site and finally inhibit tumor growth in glioma-bearing model mice [81].

4.2 BBTB-targeting strategies and related nano-drug delivery systems

With the development of brain tumors, the start of angiogenesis and gradual impairment of BBB, BBTB becomes the main obstacle for nanosystems. To date, two strategies have been proposed for BBTB targeting: i) angiogenesis-specific targeted drug delivery like Arg-Gly-Asp conserved sequence peptide (RGD) and DNA aptamer; ii) nano-drug carriers targeted drug delivery like Arg-Gly-Asp conserved sequence peptide (RGD) and DNA aptamer; iii) nano-drug carriers energy-dependent endocytosis and then intracellularly transponed involving both the Golgi apparatus and lysosomes [82]. Recently we reported a PTX-loaded cyclic RGD-poly(ethylene glycol)-block-poly(lactic acid) micelle (c(RGDyK)-PEG-PLA/PTX) drug delivery system effect. The presence of c(RGDyK) enhanced the anti-glioblastoma cell cytotoxic efficacy by 2.5-folds in the in vitro cytotoxicity study, as shown by a promising competitive binding IC$_{50}$ value as low as 26.30 nM with U87MG cells. In intracranial U87MG glioma model, c(RGDyK)-PEG-PLA/PTX realized significant glioma accumulation and prolonged median survival time (48 days for PTX-loaded c(RGDyK)-modified PEG-PLA micelle group, 41 days for PTX-loaded unmodified PEG-PLA micelle, 38 days for Taxol® and 34 days for saline) [87].

RGD-modified dendrimer PAMAM covalently conjugated to DOX with acid-sensitive cis-acioryl linkage showed an IC$_{50}$ of 3.58 µM, much lower than free DOX (8.24 µM) in human umbilical vein endothelial cells (HUVEC) [88] and preferred to accumulated at the brain tumor over nontumor tissue and prolonged the survival time of experimental glioma-bearing mice [89].

RGD also enhanced transfection efficiency of pegylated PEI as the gene carrier. Our recent report compared RGD-poly(ethylene glycol)-PEI (RGD-PEG-PEI) and RGD-PEI-methoxyl poly(ethylene glycol) (RGD-PEI-mPEG) with respect to their glioblastoma cell-binding capability and tumor-targeting ability of their complexes with plasmid DNA. RGD-PEG-PEI/plasmid-enhanced green fluorescent protein (pEGFP)-N2 complexes had higher binding affinities with U87MG cells than RGD-PEI-mPEG/pEGFP-N2 complexes and greater biodistribution of RGD-PEG-PEI/pEGFP-N2 complexes in mice bearing subcutaneous than that of RGD-PEI-mPEG/pEGFP-N2 complexes, suggesting the importance of the PEG spacer for the timely exposure of targeting moiety [90].

Another example of RGD modification is the hollow gold nanospheres (HAuNS), which could generate intense photoacoustic signals and induce efficient photothermal ablation (PTA) therapy that can be used simultaneously to image and treat cancers. Once modified by RGD, intravenous administered HAuNS led to a 20.7°C elevation in tumor temperature and a prolonged survival of tumor-bearing mice [91].

Guo et al. developed a PEG-PLGA nanoparticle drug delivery system decorated with AS1411 (Ap) as the targeting ligand to facilitate anti-glioma delivery of PTX [92]. AS1411 was a DNA aptamer that specifically binds to nucleolin, which was highly expressed in the plasma membrane of both cancer cells and endothelial cells in angiogenic blood vessels [93]. The interaction of AS1411 with nucleolin significantly enhanced cellular association of nanoparticles in C6 glioma cells and increased the cytotoxicity of its payload. Prolonged circulation and enhanced PTX accumulation at the tumor site was achieved for AS1411-modified PTX-loaded nanoparticles, which eventually obtained significantly higher tumor inhibition on mice bearing C6 glioma xenografts and prolonged animal survival on rats bearing intracranial C6 gliomas when compared with unmodified nanoparticles and free drug Taxol® [94].
4.2.2 Drug delivery aimed at the negatively charged pores of the BBB

BBB resistance to nanoparticles larger than 12 nm is believed to be constructed by ‘nanofibres’ in the luminal glycocalyx layer [95] overlaying the anatomic defects within the BBB. This negative-charged barrier has certain sensitivity to the charge of small molecules/peptides/nano-drug delivery systems particularly for long-circulating nanoparticles that circulate in vivo for a long time and can fully interact with the inner surface of microvessels. Sarin et al. conjugated rhodamine B dye to dendrimers with positively charged terminal amines that were found to induce disruption of the glycocalyx of the already porous BBB and the normally nonporous BBB and results in enhanced extravasation of rhodamine B-conjugated dendrimers across the BBB [96]. But the research on positively charged nanoparticles smaller than 12 nm for trans-BBB transport has not been expanded to drug delivery and awaits more efforts for experimental validation.

4.3 EPR effect-based strategies and related nano-drug delivery systems

Theoretically, the high resistance of BBB and BBB will limit the access of passively targeted nanovehicles to the brain tumor to the greatest extent. However, with the development of tumor, EPR effect in brain tumor tissues will also appear though much less than that for peripheral tumors, which enables nanosystems with suitable particle size to enter the brain tumor via endothelial gaps on the microvessels of brain tumors. And that may explain the successful reports of several nanocarriers without special trans-BBB transport modification.

Liposomes with suitable sizes (<100 nm), even without active-targeting modification, may be passively targeted to tumors by EPR effect. For example, Guo et al. carried out a combination therapy study of liposomal DOX (DOX-LP) and a tumor necrosis factor-related apoptosis-inducing ligand TRAIL (TRAIL-LP), to sensitize DOX on GBM cells to TRAIL-induced apoptosis. The combination therapy of TRAIL-liposome and DOX liposome displayed stronger anti-GBM effect than free drugs or liposomal drugs alone in vivo. Such synchronous systemic administration of DOX liposome and TRAIL liposome may overcome the inconvenience in the administration of chemotherapeutic agent in clinic where anticancer drugs were usually administered 24 h prior to TRAIL treatment [97].

Bernardi et al. prepared indomethacin-loaded nanocapsules [98] with poly(e-caprolactone), caprylic/capric triglyceride and sorbitan monostearate and proved its anti-glioma efficiency in an experimental glioma model [99]. The rats treated with indomethacin-loaded nanocapsules demonstrated a significant reduction in tumor size and 50% of these animals presented just cells with characteristics of a residual tumor as shown by the less intense immunostaining of nestin, which was an intermediate filament protein involved in the organization of the cytoskeleton and expressed in almost all glioblastoma multiforme. Pathological analyses showed that the treated gliomas presented a significant reduction in the mitotic index and other histological characteristics that indicated a less invasive/proliferative tumor. An important finding of the present study is that polymeric nanocapsules were able to increase the intratumoral bioavailability of indomethacin than free drug. Furthermore, indomethacin achieved a greater concentration in the hemisphere where the glioma was implanted, compared with the contralateral healthy hemisphere.

To improve the pharmacokinetics of iron oxide nanoparticles for magnetic targeting as contrast agents in magnetic resonance imaging as well as drug delivery carriers [100], Yang et al. reported a long-circulating PEG-modified, cross-linked starch MNP (PEG-MNP) suitable for magnetic targeting. By exploring the biodistribution patterns of PEG-MNPs in main organs including liver, spleen, lung and kidney, the capability of PEGylation of enhancing magnetic brain tumor targeting was proven in glioma tumors thanks to the prolonged circulation lifetime of the nanoparticles. Up to 1.0% injected dose/g tissue nanoparticle was found in the brain tumor. MRI and histological analyses visually confirmed enhanced targeting and also suggested a limited contribution of passive mechanisms to tissue retention of nanoparticles [101].

When the EPR effect exists, nano-drug delivery systems with tumor-specific targeting moiety modification may produce good brain tumor accumulation. For example, epidermal growth factor receptor (EGFR), 170-kDa transmembrane tyrosine kinase, is often overexpressed in human glioblastoma multiforme cells but undetectable or weakly expressed in normal brain [102]. So it represents an attractive molecular target for the specific delivery of therapeutic agents to high-grade gliomas where anti-EGFR antibody-modified immunoliposomes targeted to EGFR can first enter the tumor via EPR effect and then selectively enter tumor cells with the aid of EGFR. Feng et al. conjugated anti-EGF antibodies (anti-EGFR) to sodium borocaptate (BSH)-containing liposomes to deliver BSH into glioma cell with overexpressed EGFR. Immunohistochemical analysis using an anti-BSH monoclonal antibody revealed that BSH was delivered effectively into the cells but not into EGFR-deficient glioma or primary astrocytes. In an animal model of brain tumors, both the liposomes and the BSH were observed only in the tumor [103]. The effect of anti-EGFR modification on the in vitro/in vivo performance of carmustine-carrying catanionic solid lipid nanoparticles (CASLNs) has also been reported by Kuo et al. Anti-EGFR/carmustine-CASLNs demonstrated effective delivery to U87MG cells and antiproliferative efficacy against the growth of malignant brain tumors. Furthermore, the carmustine-CASLNs could selectively bind with EGFR on cells in the G0/G1 stage, inhibit the intracellular tyrosine kinase activity of EGFR, lower the EGFR expression level, then change the function of U87MG cells [104]. Cheng et al. reported an epidermal growth factor (EGF)-modified Au nanoparticle with 10-fold improved selectivity to the brain tumor compared with untargeted conjugates. This delivery system holds promise for...
future delivery of a wider range of hydrophobic therapeutic drugs for the treatment of hard-to-reach cancers [105].

5. Expert opinion: ‘whole-process targeting’ for brain tumors

The development and growth of brain tumor is a unique process with continuously changing vascular characteristics. For different development and growth stages of brain tumors, different targeting strategies and related drug delivery systems are needed. Although present brain tumor-targeted drug delivery systems have achieved significant advances in various stages of brain tumors, a series of overall targeting drug delivery strategies that we would like to call ‘whole-process targeting’ (WPT) as shown in Figure 3 are urgently needed, for more effective treatment/control of the growth and development of brain tumors, better safety of targeted drug delivery and realization of clinic objective ‘survival with tumors.’

For the early stage of brain tumor when the BBB is still intact and neovascularization has not started, BBB and the tumor cell are the targeted goal. With the development of brain tumor, the BBB becomes impaired, the BBTB constitutes the main barrier and BBTB/tumor cell-targeting is desirable. With the further development of brain tumor, EPR effect becomes significant and nanovehicles that first enter the tumor tissue via passive targeting and then be actively targeted at the tumor cells provide a good choice. Considering

---

Figure 3. ‘Whole-process targeting’ (WPT) for brain tumors.
the relative weak EPR effect for brain tumor compared with peripheral tumors, the particle size and in vivo stability of passively targeting nano-drug delivery systems should be strictly controlled.

Besides the above-mentioned targets existing in these three stages of brain tumor, cancer stem cells (CSCs) might also provide a good target. CSCs existing within tumors (e.g. glioma) possess characteristics associated with normal stem cells, specifically the ability to give rise to all cell types found in a particular cancer sample, being tumorigenic (tumor-forming) [106]. Such cells are proposed to persist in tumors as a distinct population and cause relapse and metastasis by giving rise to new tumors [107]. Therefore, development of brain CSC-specific targeting drug delivery systems holds hope for brain cancer treatment, especially for sufferers of metastatic brain tumor. However, to date no nano-drug delivery systems has been reported for brain CSC targeting. This virgin land may be a valuable opportunity and at the mean time a great challenge.

In designing WPT nano-drug delivery systems for brain, new ideas should be explored to make them ‘smart,’ that is, being able to respond to the in vivo environments. For passively targeted drug delivery systems, we can design an environmentally sensitive moiety to expose some ligand specific for tumor cell only after entering the inner tumor tissues by EPR effect. Besides, tumor cell-targeting drug with low toxicity to normal brain tissues is preferable to conventional chemotherapy, such as TRAIL, gene or proteins, p53-activating agents (small molecules or peptide just as we previously reported [108]), tyrosine kinase inhibitors and so on.

6. Conclusions

Rapid advances in the molecular neurosciences have revolutionized brain tumor-specific drug delivery with the implementation of nanotechnology, which is playing a unique and increasingly important role. We believed that whole-process targeted drug delivery systems for brain tumors designed according to characteristics of brain tumor growth will help to realize one of the most important clinic objectives ‘survival with tumors’ for brain cancer patients.

Declaration of interest

This work was supported by National Basic Research Program of China (973 Program) 2007CB935800. The authors state no conflict of interest.

Bibliography

17. Partridge WM. Crossing the blood-brain barrier: are we getting it right? Drug Discovery Today 2001;6:1-2
20. Schulingkamp RJ, Pagano TC, Hung D, et al. Insulin receptors and insulin action in the brain: review and clinical


24. Groothuis DR. The blood-brain and blood-tumor barriers: a review of strategies for increasing drug delivery. Neuro-oncol 2002;4:45-59


47. Soni S, Babbar AK, Sharma RK, et al. Delivery of hydrophobised 5-fluorouracil derivative to brain tissue through intravenous route using surface modified nanogels. J Drug Target 2006;14:87-95


Recent advances in brain tumor-targeted nano-drug delivery systems


75. Maleki S, Blakely EL, Bjorstad KA. Human glioblastoma cell lines: levels of low-density lipoprotein receptor and low density lipoprotein receptor-related protein. Cancer Res 2000;60:2300-3


106. Peter BD. Brain tumor stem cells: the cancer stem cell hypothesis writ large. Mol Oncol 2010;4:420-30

Affiliation
Yu Liu¹ & Weiyue Lu¹,²,³
¹Author for correspondence
²Fudan University, School of Pharmacy, Department of Pharmaceutics, Key Laboratory of Smart Drug Delivery (Fudan University), Ministry of Education & PLA, China
³State Key Laboratory of Molecular Engineering of Polymers (Fudan University), China
³Professor, Fudan University, School of Pharmacy, No. 826 Zhangheng Road, Shanghai 201203, China Tel: +86 21 51980006; E-mail: wylu@shmu.edu.cn