International Journal of PharmTech Research
CODEN (USA): IJPRIF, ISSN: 0974-4304
Vol.8, No.2, pp 280-288, 2015

Polymeric Nanoparticular Drug Delivery for Treatment of Brain Cancer -A Review

B.V. Nagarjuna Yadav, V.Ravichandiran And S.Sathesh Kumar*

Department of Pharmaceutics, School of Pharmaceutical Sciences, Vels University, Vels Institute of Science and Technology and Advanced Sciences (VISTAS), Chennai-600117, India.

Abstract: Nano drug delivery system is an emerging technology for the rational delivery of chemotherapeutic drugs in the treatment of cancer. Gliomas are primary CNS tumours with the ability to infiltrate in the healthy brain tissue. Malignant brain cancer is a catastrophic disease of morbidity and mortality in adults and is the second leading cause of cancer death in children. Conventional therapeutic approaches have been largely unsuccessful in providing long-term management of brain tumours. The blood-brain barrier (BBB) significantly hinders the passage of systemically delivered therapeutics and the brain extra cellular matrix limits the distribution and longevity of locally delivered agents. Polymeric nanoparticles represent a promising solution to these problems. This review will discuss about the physiology of BBB, mechanism of BBB in restricting the entry of molecules into the brain and polymeric nanoparticles. Moreover, in comparison to other nanocarrier systems, polymeric nano particles are generally safer and more stable; they can also be easier to prepare and offer better control over agent release which results in the delivery of therapeutic agents to the target site with high bioavailability and almost no drug loss. The details of various polymeric nano formulations reported by the researchers with anticancer drug were consolidated.
Key Words: Nanoparticles, Blood Brain Barrier (BBB), Glioma, Central Nervous System (CNS).

Introduction

Over the last few years a number of promising novel treatment approaches have been investigated including the application of inhibitors of receptor tyrosine kinases and downstream targets, immune-based therapies and anti-angiogenic agents (1). Unfortunately so far the major clinical trials in glioblastoma patients did not deliver clear clinical benefits. Systemic brain tumor therapy is seriously hampered by poor drug delivery to the brain (2)**.** The morphological and physiological characteristics of ceribrovascular endothelial cells which are tightly connected to each other and supported by glial cells make up a unique complex as blood brain barrier (BBB) (2,4)**.** Active cytotoxic compounds encapsulated into liposomes, micelles, and nanoparticles constitute novel treatment options because they can be designed to facilitate entry into the brain parenchyma.

Tumors of the central nervous system (CNS) are classified based on the presumed tissue of origin, i.e. tumors of neuroepithelial origin, tumors of cranial and paraspinal nerves, tumors of the meninges, lymphomas and hematopoietic neoplasms, germ cell tumors, tumor of the sellar region and metastatic brain tumors (3)**.** The majorities of malignant brain tumors in adults are of neuroepithelial origin and belong to the group of gliomas, based on their resemblance to glial support cells of the brain, astrocytes and oligodendrocytes. Glial tumors are further classified in grades (I to IV) according to their clinical manifestation and malignancy. Except for grade I pilocytic astrocytomas, all other glial tumors eventually develop into a fatal tumor albeit with different

incubation times. All these tumors are thus considered malignant. Diffusely infiltrating gliomas (grade II) mostly affect young adults with a high degree of cellular differentiation and slow growth. Over time these tumors evolve to anaplastic astrocytomas or oligodendrogliomas (grade III) or to glioblastomas (GBM). Grade IV astrocytoma or GBM represents the most malignant type of brain tumor in adults and is also the most frequently occurring primary brain tumor. Despite an aggressive treatment regimen, the median time from diagnosis to death for GBM patients is only 14 months (1).

Nanoparticles are emerging technologies for the rational delivery of chemotherapeutic drugs in the treatment of cancer (4). The use of nanoparticles release improved pharmacokinetic properties controlled and sustained release of drugs and, more importantly, lower systemic toxicity (5).

Numerous investigations have shown that both tissue and cell distribution profiles of anticancer drugs can be controlled by their entrapment in submicronic colloidal systems (nanoparticles). The rationale behind this approach is to increase antitumor efficacy, while reducing systemic side-effects (6). This review provides an update of tumor targeting with conventional or long-circulating nanoparticles. The in vivo fate of these systems, after intravascular or tumoral administration, is discussed, as well as the mechanism involved in tumor regression. Nanoparticles are also of benefit for the selective delivery of oligonucleotides to tumor cells. Moreover, certain types of nanoparticles showed some interesting capacity to reverse Multi Drug Resistance, which is a major problem in chemotherapy (7).

According to the World Health Organization (WHO) classification of the central nervous system (CNS) tumors in 2007, malignant gliomas are classified as neuroepithelial tumors. Based on this classification, these tumors can be subdivided as astrocytic, oligodendroglial, oligoastrocytic, and ependymal, on the basis of their resemblance to the glial cells (8). Gliomas are primary CNS tumors with the ability to infiltrate in the healthy brain tissue and form satellite tumors (9).

Malignant brain cancer is a catastrophic disease of morbidity and mortality in adults and is the second leading cause of cancer death in children. Malignant brain tumors are a significant health problem in children and adults. Conventional therapeutic approaches have been largely unsuccessful in providing long-term management (10). As primarily a metabolic disease, malignant brain cancer can be managed through changes in metabolic environment. In contrast to normal neurons and glia, which readily transition to ketone bodies (β-hydroxybutyrate) for energy under reduced glucose, malignant brain tumors are strongly dependent on glycolysis for energy (11).

Blood-Brain-Barrier

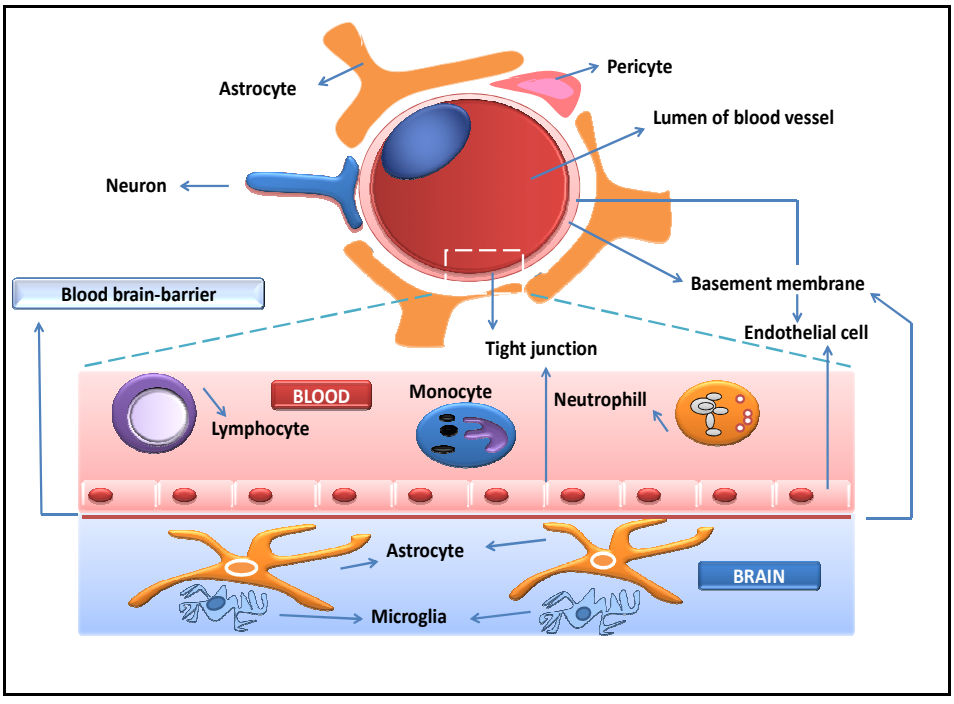

Fig 1. Schematic representation of blood brain barrier

The brain is well protected and dynamically regulated to provide a sanctuary for the central nervous system (CNS). There are several gateways to enter brain parenchyma; the most important two are blood circulation and cerebrospinal fluid (CSF) circulation (12). In the human brain, there are about 100 billion capillaries in total, providing a combined length of brain capillary endothelium of approximately 650 km and a total surface area of approximately 20 m^2 (13).

The blood–brain barrier (BBB) is a dynamic barrier protecting the brain against invading organisms and unwanted substances. It is also the most important barrier impeding drug transport into the brain via the blood circulation (14). Despite the rapid development in our understanding of the molecular structure of components of the BBB, our knowledge in receptor expression at the BBB, advances in medical technology, and breakthroughs in nanotechnology-based approaches, many of the brain or CNS associated diseases remain under-treated by effective therapies. This is not because there is a lack of candidate drugs but due to the inability of many therapeutic molecules to cross the BBB, the blood–cerebrospinal fluid barrier (BCSFB), or other specialized CNS barriers to reach the specific areas of brain (15)**.**

Any molecules' entry into the brain via parenteral administration is strictly controlled by the BBB and the BCSFB. As the surface of BCSFB faces the ventricle that is filled with CSF, not the blood (16)**.** This, in combination with the high turnover rate of CSF, leads to continuously flushing the injected drug (i.e. those injected into the ventricle) back to the blood (17). The structure of the BBB is depicted in the figure 1.

Fig 1. Schematic representation of blood brain barrier

One of the most significant technologies adopted for the localized delivery of drugs is the automatic bypass of the BBB, which results in the delivery of the therapeutic agents to the target site with high bioavailability and almost no drug loss. Various localized drug delivery approaches such as the injection and infusion of the therapeutic agents as well as the convection enhanced drug delivery (CED), and administration of implants have been introduced so far (18).

Thus, the main routes for transport across the BBB are:

- The paracellular aqueous pathway, which is restricted by the tight junctions.
- The transcellular lipophilic pathway.
- Substrate-specific transport proteins.
- Receptor-mediated transcytosis.
- Adsorptive-mediated transcytosis.

Due to the specificity and restrictive nature of the BBB, only lipophilic drugs with a molecular weight < 500 Da cross the BBB in pharmacologically significant amounts (29-33).

Nanotechnology - A Novel Drug Delivery System To Brain:

Nanoparticles are polymeric solid colloidal particles ranging in size from 10-1000 nm and are employed to carry the drugs through absorption or incorporation. The mechanism of nanoparticles mediated transport of drugs is mostly endocytosis by endothelial cells lining the brain blood capillaries.

Higher concentration gradient of drug at the blood brain barrier that may enhance the transport across the endothelial cell layer increases retention in the brain which facilitates the solubilisation of endothelial cell membrane lipids by surfactant action of nanoparticles leading to membrane fluidization and enhanced drug permeability to BBB. Loosening of tight junctions between endothelial cells and increased permeability of drug or drug-nanoparticles passes through these channels.

The earlier experiments show that detergents can cause solvent mediated BBB disruption. In order to make that possible it is necessary to administer relatively large doses of polysorbates 80, up to 200mg/kg intravenously to stabilize the nanoparticles. Recent studies show that this detergent present in formulation is responsible for enhanced BBB transport of drug/nanoparticles/Tween 80 complexes.

Since the usefulness of conventional nanoparticles is limited by their massive capture by the macrophages of the MPS after intravenous administration, other nanoparticulate devices must be considered to target tumors.

Nanoparticles for pharmaceutical and medical application are around now for over 35 years. Now there are numerous reports and studies conducted every year and their number is increasing exponentially. The first commercial nanoparticles product containing a drug (AbraxaneTM, human serum albumin nanoparticles containing paclitaxel) appeared on the market at the beginning of 2005. Nanoparticles for diagnostic purposes have been marketed now for over 10 years. A second product based on poly(isohexyl cyanoacrylate) nanoparticles (Doxorubicin-Transdrug®) loaded with doxorubicin is presently being developed by the company BioAlliance in Paris for the treatment of resistant hepatocellular carcinomas and a Phase I/II clinical trial has been conducted (18,19).

Numerous investigations have shown that both tissue and cell distribution profiles of anticancer drugs can be controlled by their entrapment in submicronic colloidal systems (nanoparticles). The rationale behind this approach is to increase antitumor efficacy, while reducing systemic side-effects.

Tumor blood vessels present several abnormalities in comparison with normal physiological vessels, often including a relatively high proportion of proliferating endothelial cells, an increased tortuosity, a deficiency in pericytes and an aberrant basement membrane formation (20). The resulting enhanced permeability of tumor vasculature is thought to be regulated by various mediators, such as vascular endothelium growth factor (VEGF), bradykinin, nitric oxide, prostaglandins and matrix metalloproteinases.

A strategy could be to associate antitumor drugs with colloidal nanoparticles, with the aim to overcome non-cellular and cellular based mechanisms of resistance and to increase selectivity of drugs towards cancer cells while reducing their toxicity towards normal tissues (21).

According to the process used for the preparation of the nanoparticles, nanospheres or nanocapsules can be obtained. Unlike nanospheres (matrix systems in which the drug is dispersed throughout the particles), nanocapsules are vesicular systems in which the drug is confined to an aqueous or oily cavity surrounded by a single polymeric membrane (22,23). Nanocapsules may, thus, be considered as a 'reservoir' system if designed appropriately; nanoparticles may act as a drug vehicle able to target tumor tissues or cells, to a certain extent, while protecting the drug from premature inactivation during its transport. Indeed, at the tumor level, the accumulation mechanism of intravenously injected nanoparticles relies on a passive diffusion or convection across the leaky, hyper permeable tumor vasculature. The uptake can also result from a specific recognition in case of ligand decorated nanoparticles ('active targeting') (24).

Polymeric nanoparticles have been shown to be characterized by a prolonged half-life in the blood compartment (25). This allows them to selectively extravasate in pathological sites, like tumors or inflamed regions with a leaky vasculature. As a result, such long-circulating nanoparticles are supposed to be able to directly target most tumors located outside the MPS regions. The size of the colloidal carriers as well as their surface characteristics are the key for the biological fate of nanoparticles, since these parameters can prevent their uptake by MPS macrophages (26).

The use of hydrophilic polymers like poly(ethylene glycol(PEG), poloxamines, poloxamers, polysaccharides provided a new dimension for the application of nanoparticles to efficiently coat conventional nanoparticles surface. These coatings provide a dynamic 'cloud' of hydrophilic and neutral chains at the particle surface, which repel plasma proteins, as modeled by Jeon et al. Hydrophilic polymers can be introduced at the surface in two ways, either by adsorption of surfactants or by use of block or branched copolymers. Coating conventional nanoparticles with surfactants, in order to obtain a long-circulating carrier, has been the first strategy used to direct tumor targeting in vivo (27).

The blood–brain barrier significantly hinders the passage of systemically delivered therapeutics and the brain extracellular matrix limits the distribution and longevity of locally delivered agents. Polymeric nanoparticles represent a promising solution to these problems. Over the past 40 years, substantial research efforts have demonstrated that polymeric nanoparticles can be engineered for effective systemic and local delivery of therapeutics to the CNS. Moreover, many of the polymers used in nanoparticle fabrication are both biodegradable and biocompatible, thereby increasing the clinical utility of this strategy (28).

The polymeric nanoparticles are stable and allow high loading of many agents, they provide control over drug release kinetics, they can be readily modified to display a variety of surface-attached ligands, and many polymers have a long history of safe use in humans (29).

In general, for optimal CNS delivery, nanoparticles should be:

- Scalable and cost-effective,
- Biocompatible/ biodegradable,
- Non-toxic,
- Non-immunogenic,
- Below100 nm in diameter,
- Amenable to robust surface modification.

Many natural and synthetic polymers have been used to create nanoparticles for CNS delivery, including polysaccharides, proteins, amino acids, poly(ethylenimines), poly (alkylcyanoacrylates), poly(-methylidene malonates), and polyesters. Systemic delivery of polymeric nanoparticles to the CNS is based largely on their potential for receptor-mediated transcytosis and adsorptive-mediated transcytosis through the BBB. This process can be enhanced by the addition of cell-penetrating peptides and/or targeting ligands to the nanoparticle surface. In studies to date, the nanoparticle systems described in this section have shown the most promise for bypassing the BBB.

Chitosan nanoparticles are produced by electrostatic interactions between positively charged chitosan and a polyanion sodium tripolyphosphate. PEG-grafted chitosan nanoparticles have been also produced in order to improve their long-circulating properties. Nevertheless, their large size, between 200nm and 1 mm, is a potential disadvantage for the delivery of drugs to the brain. Coacervation methods are also used to prepare chitosan or gelatin nanoparticles (30).

The first nanoparticulate systems that were used for direct drug delivery to the brain were microspheres. Polymer microspheres have been fabricated from a variety of materials for the purposes of local delivery including PLGA, poly (methylidene malonate) (PMM), poly (epsiloncaprolactone), and chitosan (31,32).

These systems have been used to deliver a range of therapeutics, including cyclosporine, paclitaxel, imatinib, mitoxantrone, phenytoin, and nerve growth factor. One advantage of microparticles, over earlier implant systems such as Gliadel®, is that the particles can be introduced without surgery (33,34). But, because particles larger than 1 micron in diameter do not move readily through the BBB or the brain interstitium, it is difficult formicroparticles to distribute through large volumes of brain tissue (35). In contrast, when nanoparticles are used to deliver agents instead of microparticles, particularly nanoparticles that are less than 100 nm in diameter, CED can be used to transport the particles over clinically relevant volumes of distribution (36-37).

Although several clinical trials have investigated the role of nanoliposomal vehicles for CNS drug delivery, there have not yet been similar studies for polymeric nanoparticles (38).

Moreover, in comparison to other nanocarrier systems, polymeric nanoparticles are generally safer and more stable; they can also be easier to prepare and offer better control over agent release. As this technology moves forward, some of the major challenges to clinical translation will be the ability to scale-up this system in a cost-effective manner. The aging population and increasing prevalence of neurological disorders, the demand for improved CNS therapeutics is only going to increase with time. In particular, the application of polymeric nanoparticles to CNS malignancies, neurodegenerative disorders, and ischemic disease will be of interest (39). The formulation technologies adopted for the development of anticancer loaded polymeric nanoparticles were listed in the table 1.

Table 1: polymeric nanoparticles loaded with anticancer drugs

S. No	Name of the Anticancer Drug	Polymer	Surfactant Used	Method of preparation of Nano Formulations	Animal Model	Cell lines used	References
1	Paclitaxel	Methoxyl poly(ethylene glycol), PCL	-	O/W Emulsion and Evaporation Technique (Nanoparticles)	Male ICR mice	C6 cells	39
2	Dopamine	Chitosan	-	Modified Ionic Gelation Method (Magnetic Nanoparticles)	Male Wistar rats	MDCKII-MDR1 cells	40

3	Camptothecin	PLGA	-	Nanoprecipitation Method (Nanoparticles)	Syngeneic Fischer F344 Female rats	Rat gliosarcoma 9L cells.	41
4	Paclitaxel	PEGylated poly(trimethylene carbonate)	-	Emulsion/Solvent Evaporation Technique (Nanoparticles)	Balb/c nude mice	U87MG cells	42
5	Camptothecin	Poly(DL-lactic acid) (PLA)	-	O/W emulsification and Subsequent evaporation of organic solvent. (Nanoparticles)	Male Wistar rats and male ddY mice	Sarcoma 180 (S-180) cells balanced solution (0.1 ml) per mouse	43
6	Teniposide	PLGA	-	Modified Oil-In-Water (O/W) Single-Emulsion Solvent Evaporation Process (Nanoparticles)	----------	U87MG cells	44
7	Doxorubicin	Poly(isohexyl cyanoacrylate)	-	Anionic Emulsion Polymerization (Nanoparticles)	Male Wistar rats	Glioblastoma cell line 101/8	45
8	Paclitaxel	PLGA	-	Emulsion/Solvent Evaporation Method (Nanoparticles)	SD rats (200 ±10 g)	C6 cells	46
9	Penetratin Coumarin	MePEG–PLA	-	emulsion/solvent evaporation technique	Sprague-Dawley rats	One hundred microliters of MDCK–MDR cells	47
10	Temozolomide	PLA	-	solvent evaporation method	--	C6 glioma cell	48
11	Gemcitabine	PBCA	Polysorbate 80	Emulsion polymerization method	Sprague Dawley (SD) rats	C6 glioma cells	49
12	Paclitaxel	Methoxyl poly(ethylene glycol)	-	O/W emulsion and evaporation technique	Male BALB/c nude mice (20±2 g) and ICR mice (20±2 g)	C6 cells	50
13	Rivastigmine	Chitosan	-	Ionic gelation method	Wistar rats (aged, 4–5 months)	Nose-to-brain delivery of placebo nanoparticles (CSNPs) was investigated by confocal laser scanning microscopy technique using rhodamine-123 as a marker.	51

14	Tacrine	PBCA	Polysorbate 80	Emulsion polymerization method (Nanoparticles)	Wistar rats 180–220 g	--	52
15	Temozolomide	PLGA	Polysorbate 80	emulsifying-solvent evaporation method super paramagnetic nanoparticles	-	Glioma C6 cell lines	53

Conclusion

With a high degree of constraints and with a greater demand for transport of anticancer drugs into brain, the complication of a safe and effective drug delivery system is struggling. The nanoparticles is seen as a ray of hope for the pharmaceutical formulation scientists as they had the well proved the efficiency to transport the drugs across the BBB and deliver them with high degree of specificity to the target site. Further the polymeric nanoparticles can be deliberately moved to the required receptors by performing necessary surface engineering. No doubt that the polymeric nanoparticles are going to be one of the major breakthroughs in chemotherapy of brain cancer.

References

1. Swapnil V, Bhujbal, Paul de Vos, Simone P, Niclou, drug and cell encapsulation: Alternative delivery options for the treatment of malignant brain tumors, Advanced Drug Delivery Reviews Accepted 23 January 2014.
2. Takayoshi Yoshikawaa, Toshiyuki Sakaeda (nee Kakutani), Tamio Sugawara Koichiro Hirano, Valentino J. Stella A novel chemical delivery system for brain targeting Advanced Drug Delivery Reviews 36 (1999) 255–275.
3. G.W. Goldstein, A.L. Betz, The blood–brain barrier, Sci. Am. 255 (1986) 70–79.
4. K.L. Audus, P.J. Chikhale, D.W. Miller, S.E. Thompson, R.T. Borchardt, Brain uptake of drugs: The influence of chemical and biological factors, Adv. Drug Res. 23 (1992) 1–64.
5. R. Stupp,W.P.Mason, M.J. van den Bent,M.Weller, B. Fisher,M.J.B. Taphoorn, et al. Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma, N. Engl. J. Med. 352 (2005) 987–996.
6. Yogeshkumar Malam, Marilena Loizidou and Alexander M. Seifalian, Liposomes and nanoparticles: nanosized vehicles for drug delivery in cancer, Trends in Pharmacological Sciences Vol.30 No.11.
7. Irène Brigger, Catherine Dubernet, Patrick Couvreur, Nanoparticles in cancer therapy and diagnosis, Advanced Drug Delivery Reviews 64 (2012) 24–36.
8. Daniela Allhenna, Maryam Alsadat Shetab Boushehria, Alf Lamprechta, Drug delivery strategies for the treatment of malignant gliomas International Journal of Pharmaceutics 436 (2012) 299– 310.
9. American Cancer Society, Cancer Facts & Figures 2009, American Cancer Society, Atlanta, 2009, p. 68.
10. R.E. McLendon, E.C. Halperin, Is the long-term survival of patients with intracranial glioblastoma multiforme overstated? Cancer 98 (2003) 1745–1748.
11. P.J. Jukich, B.J. McCarthy, T.S. Surawicz, S. Freels, F.G. Davis, Trends in incidence of primary brain tumors in the United States, 1985–1994, Neuro-oncology 3 (2001) 141–151.
12. P. Kaatsch, C.H. Rickert, J. Kuhl, J. Schuz, J. Michaelis, Population-based epidemiologic data on brain tumors in German children, Cancer 92 (2001) 3155–3164.
13. Thomas N. Seyfried , Michael A. Kiebish , Jeremy Marsh , Laura M. Shelton, Leanne C. Huysentruyt , Purna Mukherjee, Metabolic management of brain cancer, Biochimica et Biophysica Acta 1807 (2011) 577–594.
14. Yan Chen, Lihong Liu Modern methods for delivery of drugs across the blood–brain barrier Advanced Drug Delivery Reviews 64 (2012) 640–665.
15. W.M. Pardridge, Blood–brain barrier drug targeting: the future of brain drug development, Mol. Interv. 3 (2003) 90–105 151.

16. J. Rip, G.J. Schenk, A.G. de Boer, Differential receptor-mediated drug targeting to the diseased brain, Expert Opin. Drug Deliv. 6 (2009) 227–237.
17. S.A. Pathan, Z. Iqbal, S.M. Zaidi, S. Talegaonkar, D. Vohra, G.K. Jain, A. Azeem, N. Jain, J.R. Lalani, R.K. Khar, F.J. Ahmad, CNS drug delivery systems: novel approaches, Recent Pat. Drug Deliv. Formul. 3 (2009) 71–89.
18. Paolo Blasi, Stefano Giovagnoli, Aurélie Schoubben, Maurizio Ricci, Carlo Rossi Solid lipid nanoparticles for targeted brain drug delivery Advanced Drug Delivery Reviews 59 (2007) 454–477.
19. L.W. Seymour, Passive tumor targeting of soluble macromolecules and drug conjugates, Crit. Rev. Ther. Drug Carrier Syst. 9 (1992) 135–187.
20. D. Baban, L.W. Seymour, Control of tumor vascular permeability, Adv. Drug Deliv. Rev. 34 (1998) 109–119.
21. H. Maeda, The enhanced permeability and retention (EPR) effect in tumor vasculature: the key role of tumor-selectivemacromolecular drug targeting, Adv. Enzyme Regul. 41 (2001) 189–207.
22. P. Couvreur, G. Couarraze, J.-P. Devissaguet, F. Puisieux, Nanoparticles: preparation and characterization, in: S. Benita (Ed.), Microencapsulation: Methods and Industrial Application, Marcel Dekker, New York, 1996, pp. 183–211.
23. F. Yuan, Transvascular drug delivery in solid tumors, Semin. Radiat. Oncol. 8 (1998) 164–175.
24. S.M. Moghimi, A.C. Hunter, J.C. Murray, Long-circulating and target-specific nanoparticles: theory to practice, Pharmacol. Rev. 53 (2001) 283–318.
25. Irène Brigger, Catherine Dubernet, Patrick Couvreur Nanoparticles in cancer therapy and diagnosis Advanced Drug Delivery Reviews 64 (2012) 24–36.
26. S. Stolnik, L. Illum, S.S. Davis, Long circulating microparticulate drug carriers, Adv. Drug Deliv. Rev. 16 (1995) 195–214.
27. R. Gref, Y.Minamitake,M.T. Peracchia, V. Trubetskoy, V. Torchilin, R. Langer, Biodegradable long-circulating polymeric nanospheres, Science 263 (1994) 1600–1603.
28. Toral Patel, Jiangbing Zhou, Joseph M. Piepmeier, W. Mark Saltzman, Polymeric nanoparticles for drug delivery to the central nervous system, Advanced Drug Delivery Reviews 64 (2012) 701–705.
29. W.M. Saltzman, Drug Delivery: Engineering principles for drug therapy, Oxford University Press, New York, 2001.
30. Sarmento B, Martins S, Ribeiro A, Veiga F, Neufeld R, Ferreira D. Development and comparison of different nanoparticulate polyelectrolyte complexes as insulin carriers. Int J Pept Res Ther 2006; 12(2):131–8.
31. O. Benny, L.G.Menon, G. Ariel, E. Goren, S.K. Kim, C. Stewman, P.M. Black, R.S. Carroll, M. Machluf, Local delivery of poly lactic-co-glycolic acid microspheres containing imatinib mesylate inhibits intracranial xenograft glioma growth, Clin. Cancer Res. 15 (2009) 1222–1231.
32. E. Fournier, C. Passirani, C. Montero-Menei, N. Colin, P. Breton, S. Sagodira, P. Menei, J.P. Benoit, Therapeutic effectiveness of novel 5-fluorouracil-loaded poly (-methylidene malonate 2.1.2)-based microspheres on F98 glioma-bearing rats, Cancer 97 (2003) 2822–2829.Z.
33. Li, Q. Li, S. Simon, N. Guven, K. Borges, B.B. Youan, Formulation of spray-dried phenytoin loaded poly(epsilon-caprolactone) microcarrier intended for brain delivery to treat epilepsy, J. Pharm. Sci. 96 (2007) 1018–1030.
34. O.F. Turkoglu, H. Eroglu, O. Gurcan, E. Bodur, M.F. Sargon, L. Oner, E. Beskonakli, Local administration of chitosan microspheres after traumatic brain injury in rats: a new challenge for cyclosporine–a delivery, Br. J. Neurosurg. 24 (2010) 578–583.
35. M. Yemisci, S. Bozdag,M. Cetin, F. Soylemezoglu, Y. Capan, T. Dalkara, I. Vural, Treatment of malignant gliomas with mitoxantrone-loaded poly (lactide-co-glycolide) microspheres, Neurosurgery 59 (2006) 1296–1302.
36. W.M. Saltzman, M.W. Mak, M.J. Mahoney, E.T. Duenas, J.L. Cleland, Intracranial delivery of recombinant nerve growth factor: release kinetics and protein distribution for three deliveery systems, Pharm. Res. 16 (1999) 232–240.
37. S.K. Hobbs, W.L. Monsky, F. Yuan, W.G. Roberts, L. Griffith, V.P. Torchilin, R.K. Jain, Regulation of transport pathways in tumor vessels: role of tumor type and microenvironment, Proc. Natl. Acad. Sci. U. S. A. 95 (1998) 4607–4612.
38. R.G.Thorne, C. Nicholson, In vivo diffusion analysis with quantum dots and dextrans predicts the width of brain extracellular space, Proc. Natl. Acad. Sci. U. S. A. 103 (2006) 5567–5572.
39. Hongliang Xin, Liangcen Chen, Jijin Gu, Xiaoqing Ren, Zhang wei, Jieqi Luo, Yanzuo Chen, Xinyi Jiang, Xianyi Sha, Xiaoling Fang. Enhanced anti-glioblastoma efficacy by PTX-loaded PEGylated poly

(_-caprolactone) nanoparticles: In vitro and in vivo evaluation International Journal of Pharmaceutics 402 (2010) 238–247.

40. Adriana Trapania, Elvira De Giglio, Damiana Cafagna, Nunzio Denora, Gennaro Agrimi, Tommaso Cassano, Silvana Gaetani, Vincenzo Cuomo, Giuseppe Trapani Characterization and evaluation of chitosan nanoparticles for dopamine brain Delivery International Journal of Pharmaceutics 419 (2011) 296– 307.
41. Yasemin C, ırpanlıa , Emilie Allard, Catherine Passirani, Erem Bilensoy, Laurent Lemaire, Sema C, alıs Jean-Pierre Benoit Antitumoral activity of camptothecin-loaded nanoparticles in 9L rat glioma modelInternational Journal of Pharmaceutics 403 (2011) 201–206.
42. Xinyi Jiang, Hongliang Xin, Xianyi Sha, Jijin Gu, Ye Jiang, Kitki Law, Yanzuo Chen, Liangcen Chen, Xiao Wang, Xiaoling Fang PEGylated poly(trimethylene carbonate) nanoparticles loaded with paclitaxel for the treatment of advanced glioma: In vitro and in vivo evaluation International Journal of Pharmaceutics 420 (2011) 385– 394.
43. Ryotaro Kunii, Hiraku Onishi, Yoshiharu Machida Preparation and antitumor characteristics of PLA/(PEG-PPG-PEG) nanoparticles loaded with camptothecin European Journal of Pharmaceutics and Biopharmaceutics 67 (2007) 9–17.
44. Liqian Mo, Lianbing Hou, Dan Guo, Xiaoyan Xiao, Ping Mao, Xixiao Yang Preparation and characterization of teniposide PLGA nanoparticles and their uptake in human glioblastoma U87MG cells international joural of pharmaceutics 436 (2012) 812-824.
45. Stefanie Wohlfart, Alexander S. Khalansky, Christian Bernreuther Martin Michaelis, Jindrich Cinatl Jr, Markus Glatzel, Jörg Kreuter Treatment of glioblastoma with poly(isohexyl cyanoacrylate) nanoparticles International Journal of Pharmaceutics 415 (2011) 244– 251.
46. Jianwei Guo, Xiaoling Gao, Lina Su, Huimin Xia, Guangzhi Gu, Zhiqing Pang, Xinguo Jiang,Lei Yao, Jun Chen , Hongzhuan Chen Aptamer-functionalized PEGePLGA nanoparticles for enhanced anti-glioma drug delivery Biomaterials 32 (2011) 8010-8020.
47. Huimin Xia, Xiaoling Gao, Guangzhi Gu, Zhongyang Liu, Quanyin Hu, Yifan Tu,Qingxiang Song, Lei Yao, Zhiqing Pang, Xinguo Jiang, Jun Chen, Hongzhuan Chen Penetratin-functionalized PEG–PLA nanoparticles for brain drug delivery International Journal of Pharmaceutics 436 (2012) 840– 850.
48. Darshana S. Jain, Rajani B. Athawale, Amrita N. Bajaj, Shruti S. Shrikhande,Peeyush N. Goel, Yuvraj Nikam, Rajiv P. Gude Poly lactic acid (PLA) nanoparticles sustain the cytotoxic action of temozolomide in C6 Glioma cells Biomedicine & Aging Pathology 3 (2013) 201–208.
49. Chun-Xia Wang, Le-Song Huang, Lian-Bing Hou, Liang Jiang, Zhi-Ting Yan,Yu-Lun Wang, Zhi-Liang Chen Antitumor effects of polysorbate-80 coated gemcitabine polybutylcyanoacrylate nanoparticles in vitro and its pharmacodynamics in vivo on C6 glioma cells of a brain tumor model Brain Research 1261 (2009) 91-99.
50. Hongliang Xin, Liangcen Chen, Jijin Gu, Xiaoqing Ren, Zhang wei, Jieqi Luo,Yanzuo Chen, Xinyi Jiang, Xianyi Sha, Xiaoling Fang Enhanced anti-glioblastoma efficacy by PTX-loaded PEGylated poly(_-caprolactone) nanoparticles: In vitro and in vivo evaluation International Journal of Pharmaceutics 402 (2010) 238–247.
51. Mohammad Fazil, Shadab Md, Shadabul Haque, Manish Kumar, Sanjula Baboota , Jasjeet kaur Sahni, Javed Ali Development and evaluation of rivastigmine loaded chitosan nanoparticles for brain targeting European Journal of Pharmaceutical Sciences 47 (2012) 6–15.
52. Barnabas Wilson, Malay Kumar Samanta a, Kumaraswamy Santhi,Kokilampal Perumal Sampath Kumar, Nallupillai Paramakrishnan, Bhojraj Suresh Targeted delivery of tacrine into the brain with polysorbate 80-coated poly(n butylcyanoacrylate) nanoparticles European Journal of Pharmaceutics and Biopharmaceutics 70 (2008) 75–84.
53. You Linga, Kun Weia, Fen Zoua, Shizheng Zhonga, Temozolomide loaded PLGA-based superparamagnetic nanoparticles for magnetic resonance imaging and treatment of malignant glioma International Journal of Pharmaceutics 430 (2012) 266– 275.

*****