Abstract
The aim of any drug delivery system is not only to deliver a drug to specific site of action but also to maintain its therapeutic concentration at the targeted site. Most of the drugs used in CNS disorders cannot cross the blood brain barrier (BBB) due to their large molecular size, less lipid solubility and p-glycoprotein (p-gp) efflux mechanism resulting in low drug concentration in brain. Among the current strategies for brain targeting drug delivery, biodegradable polymeric nanoparticles are significant in delimiting the blood brain barrier, increasing the loading efficiency in brain and also reducing the peripheral toxicity. The present review emphasizes on the surface modified polymeric nanoparticles in enhancing drug delivery across the blood brain barrier.

Keywords: Blood brain barrier, polymeric nanoparticles, drug delivery, brain targeting, coated nanotechnology, ligand nanotechnology

Introduction
Despite tremendous research, the death rate of patients suffering from brain disorders like brain tumors, HIV encephalopathy, epilepsy, cerebrovascular disease, neurodegenerative disorders, are more than that dying of systemic cancer or heart disease. The failure is due to an inefficient drug delivery as drug accessibility to the Central Nervous System (CNS) is limited by the Blood Brain Barrier (BBB) and efflux transport system (1). Essential nutrients and oxygen are supplied to the brain by blood capillaries. The walls of the blood capillaries form the so called Blood Brain Barrier (BBB). A solid connection is present between the blood vessels of BBB, and is formed by special protein complexes of endothelial cells called tight junction. The abluminal side of these endothelial cells contains pericytes, a part of BBB. The pericytes are encapsulated by the basal membrane of the endothelial cells, and are responsible for the synthesis as well as release of different components of the basal membrane and the extracellular matrix such as collagen and glycosaminoglycan. Pericytes maintains the stability of the blood vessel and also the functioning of BBB. Another type of endothelial cell is the astrocyte responsible for the hoemeostatis and the ion regulation in the brain (2). Their endfeets attach to the pericytes and the endothelial cells, covering partially the blood vessels but are not connected to other cells by tight junction (3). Astrocytes allow polar molecules entry into the nerve fluid; while pericytes eradicate the entry of polar molecules through the BBB.

Several mechanisms like passive transport, active transport, receptor mediated transport, endocytosis or transcytosis are followed by several substances to cross the BBB. These are called influx transport system, allowing the entry of essential substances from the blood into the BBB (4). The influx transport system across the BBB describes as passive transport and active transport. Passive transport allows the influx of substances having good lipophilicity, less protein...
binding and low molecular weight. The active transport includes transporter mediated transcytosis and receptor mediated endocytosis (5). Transporter mediated transcytosis is responsible for transport of small hydrophilic molecules such as amino acid, glucose and other molecules through the transporters present at the luminal and abluminal side of the endothelial cells. Receptor mediated endocytosis is responsible for transport of large or hydrophilic essential molecules such as hormones, transferrin or iron, insulin and lipoproteins by acting on receptors located on the luminal side of the endothelial cells.

On the contrary is the efflux transport system of P-glycoprotein (Pgp), multidrug resistance protein (MRP) forcing the inverse movement of many substances from the cerebral parenchyma to the blood (6). Thus the tight junction in BBB, efflux transport system restricts entry of most of drugs making many drug based therapy inefficient such as antibiotics, antiviral drugs, antiretroviral drugs etc. The lack of essential characteristics in most drugs like lipid solubility, low molecular size prevents their ability to cross BBB. Some of the large sized molecules like oligonucleotides, antibodies, peptides, proteins are out of reaching BBB (7). Several strategies are followed to overcome these barriers in order to have an efficient brain delivery of drugs. Among the several strategies, nanoparticles are considered as the best to carry drugs across the Blood brain barrier (BBB).

Nanoparticles satisfies many of the characteristics of the magic bullet concept as carrier and also when coated with ligands. Nanoparticles are colloidal matrix of natural/synthetic polymers ranging in size between 10 and 1000 nm (8). The drugs may be adsorbed to the surface of nanoparticles or entrapped within the matrix. Among the several varieties of nanoparticles polymeric nanoparticles are significant in brain targeted drug delivery due to advantages as they are inert, biocompatible and biodegradable. The smaller size of polymeric nanoparticles (< 100 nm) enables it to cross the BBB. Both hydrophilic and hydrophobic drugs can be delivered across the BBB. They are easily processed, nontoxic, and nonantigenic and also are easily delivered through blood capillaries. They protect the drugs against degradation. They are target specific drug delivery with sustained release behavior (7, 9). Both synthetic and natural polymers can be used for preparing polymeric nanoparticles. Polymers for preparing nanoparticles (10) can be classified as shown in table 2.

### Biodistribution of nanoparticles in body

After intravenous administration, polymeric nanoparticles come in contact with plasma/serum proteins before reaching the target cells. The interaction of polymeric nanoparticles with phagocytes is regulated by the balance between two serum components – opsonin which promotes phagocytosis and dysopsonin which suppress the process. The opsonin gets adsorbed to the surface of polymeric nanoparticles and makes it recognisable to the reticuloendothelial cells (RES) (11, 12). Following intravenous administration, polymeric nanoparticles are taken rapidly by RES present in liver, spleen, bone marrow and distributed rapidly into the liver (60-90) % and spleen (2-10) % and to a minor degree into the bone marrow (13). A low concentration

<table>
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<tr>
<th>No.</th>
<th>Classification of polymers</th>
<th>Examples</th>
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</thead>
<tbody>
<tr>
<td>1</td>
<td>Natural biodegradable polymers</td>
<td>Alginates, Chitosan, Gelatin, Pellula, Gliadin</td>
</tr>
<tr>
<td>2</td>
<td>Synthetic biodegradable polymers</td>
<td>PLA, PGA, PLGA, Polyethyleneglycol, Polyglycolic acid, Polyethylcyanoacrylate</td>
</tr>
<tr>
<td>3</td>
<td>Nonbiodegradable polymers</td>
<td>Polymethylmethacrylate(PMMA), Polymethylacrylate (PMA)</td>
</tr>
</tbody>
</table>
of nanoparticles can enter brain due to their uptake by RES following intravenous administration. Several technologies based on surface modification of nanoparticles are worked out to overcome the problems in connection with phagocytosis so as to enhance the concentration of drug in the brain.

**Approaches for surface modification of nanoparticles**

**Coated nanotechnology:** Coated nanotechnology is based on specialized coating of nanoparticles using polymers or surfactants which allow mimicking the molecules that would be normally transported into the brain (5). The coating of nanoparticles is done by Incubation method. In this method, the coating solution is added to the preformed nanoparticle formulation and is kept for stirring or overnight incubation is done. The coating materials for polymeric nanoparticles are discussed below.

**Polysorbate 80:** Several drugs are being reported to be successfully delivered to brain using polysorbate 80 as coating material. The coating of nanoparticles by polysorbate 80 is done by adding polysorbate 80 (1% v/v) to the already prepared drug loaded polymeric nanoparticles and kept under stirring for 30 minutes (14). It is also reported that after the addition of polysorbate 80 (1% v/v) to a model drug loaded polymeric nanoparticles, it was stored for 24 hrs (15). Dalargin adsorbed on polybutylcyanocrylate (PBCA) nanoparticles coated with polysorbate 80 was the first compound delivered to the brain, showed positive analgesic effect in rats (16). In a study, polysorbate 80 coated Gemcitabine loaded PBCA nanoparticles; efficiently carried the drug to brain as its antitumour activity was observed on C6 glioma cells of a brain tumour model (17). An attempt was also made for the delivery of Nerve growth factor (NGF) using polysorbate 80 coated PBCA nanoparticles as carrier. NGF is needed in age related neurodegenerative diseases such as Amnesia, Parkinsonism; but entry to brain is restricted by the blood brain barrier. NGF loaded PBCA nanoparticles coated with polysorbate 80 could efficiently carry NGF to the brain as evidenced by pharmacokinetic models (18). Polysorbate 80 coated chitosan nanoparticles successfully delivered Gallic acid to brain for antidepressant activity (19). Polysorbate 80 coated nanotechnologies could also efficiently deliver Doxorubicin (20), Rivastigmine (21), Met Enkephalin Kytorphin (22) to brain.

But the mechanism behind the nanoparticles mediated transport across the BBB is yet to be fully understood. Several mechanisms were suggested – an increased retention time of the nanoparticles in the brain capillaries could enhance transport of drug across the BBB, polysorbate 80 increases the drug permeability by fluidization of brain endothelial cell membrane, opening of the brain endothelial cells tight junction by nanoparticles, endocytosis of nanoparticles by the brain endothelial cells deliver the drug into the brain, transcytosis could be possible for drug loaded nanoparticles, polysorbate 80 could inhibit the P-glycoprotein (P-gp) efflux (23). Among the several mechanisms, the most probable mechanism is endocytosis (24). PBCA nanoparticles coated with polysorbate 80 may covalently couple with apolipoprotein E, A-I or B-100 in the bloodstream. Apolipoprotein bound to the surface of PBCA nanoparticles mimics low density lipoprotein (LDL). It acts on the LDL present in the brain endothelial cells and undergoes receptor mediated endocytosis (24). Finally, the drug can be delivered by passive diffusion into the brain. However, the reported most probable mechanism suffers from several disagreements as apolipoprotein E adsorption is not only specific to polysorbate 80 coated nanoparticles surfaces but also get adsorbed onto PEGylated polylactic acid nanoparticles. Polysorbate 80 is not reported to be a good coating material for polymethylmethacrylate (PMMA) nanoparticles and polystyrene nanoparticles because polysorbate 80 coated PMMA nanoparticles are not distributed to brain after intravenous administration and also polysorbate 80 coated polystyrene nanoparticles are not able to deliver Dalargin to brain (25). It is
also reported that a desirable therapeutic concentration of drug in brain cannot be attained due to the fact that polysorbate 80 competes with proteins in blood plasma causing rapid degradation of nanoparticles in serum/plasma inducing desorption of drug adsorb onto polybutylcyanoacrylate (PBCA) nanoparticles. Thus the desorption evidence that the pharmacokinetic profile of drug in brain remains similar to drug solution administered intravenously (26). It is also reported that polysorbate 80 causes an increase in brain permeability due to BBB disturbances (27); polysorbate 80 coated nanoparticles causes BBB toxicity evidenced on the basis of sucrose permeability test (20 mg/kg in rats). Polysorbate 80 coated PBCA nanoparticles decreased locomotor activity in mice when investigated (28) and also reported that its short duration of pharmacological action needs regular intravenous administration which makes it unsuitable for chronic brain disorders. PBCA is also reported as a synergistic factor for enhancing brain permeability. In comparison to PBCA, Polylactide (PLA) or Poly (lactide-co-glycolide) (PLGA) microspheres are reported to be of good CNS biocompatibility (25).

**Glutathione:** Glutathione is considered better than Polysorbate 80 as a coating material. Unlike polysorbate 80, glutathione is an endogenous peptide and not toxic to body. Using glutathione as a coating material, an attempt has been made for the delivery of Paclitaxel across the BBB. Glutathione coated Polylactide-co-glycolide (PLGA) nanoparticles reported to be a good carrier for Paclitaxel to brain as investigated by PgpATPase assay. Glutathione is reported to act by inhibiting the Pgp efflux transport system (29).

Doxorubicin lacks permeability to brain due to its low lipophilicity, high molecular weight and efflux by Pgp. Glutathione coated on Doxorubicin adsorbed to PLGA PEG nanoparticles act against Pgp efflux transport system making Doxorubicin accessible to brain (29).

**Mannan:** Mannan coated Gelatin nanoparticles were reported as a successful carrier for Didanosine to brain. Gelatin is a biocompatible, biodegradable polymer (30). Various surface receptors like mannosyl, lectin and galactosyl present in macrophages of brain help in recognisation and endocytosis of nanoparticulate carriers. Due to this fact, nanoparticles containing ligands such as mannosyl, immunoglobulin, fibronectin and galactosyl are better phagocytosed by macrophages than carriers without such ligands. The mannan, coated on the surface of gelatin nanoparticles are recognised by mannosyl receptors present predominantly on the macrophages of brain (31) and phagocytosed by the macrophages leading to an effective delivery to brain. Mannan coating of nanoparticle suspension was done by incubation method (32) where mannansolution (1% m/v) was prepared in hot water and mixed with 1.0ml of preformed nanoparticle suspension; kept overnight stirring at room temperature (31).

**Albumin:** Albumin can be safely used as a coating material for nanoparticles as albumin coated nanoparticles in mice reported to have no mortality with upto a 2000 mg/kg (25).

**Poloxamer:** Poloxamer is considered to play a significant role in drug delivery to brain. Probable mechanism of poloxamer coated nanoparticles includes inhibition of Pgp efflux transport system and multi drug resistance protein efflux transport mechanism (33). It is also reported that apolipoproteins adsorbed on the surface of poloxamer coated nanoparticles; ligands and monoclonal antibodies conjugated to the poloxamer coated nanoparticles could cross the BBB via specific endogenous transporters localised within the brain capillary endothelium (34). Poloxamer 188 coated PBCA nanoparticles is reported to be a good carrier for Doxorubicin against an intracranial glioblastoma in rat (35). Poloxamer coating on drug loaded polymeric nanoparticles is also done by Incubation method. As an example, the coating of poloxamer on Acyclovir loaded PLGA nanoparticles was done by mixing poloxamer (1 % w/v) solution with uncoated Acyclovir loaded PLGA nanoparticles followed by overnight incubation (36)

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Polyethylene glycol (PEG): Polyethylene glycol (PEG) coating enhances half-life of nanoparticles by several magnitudes. PEG coating provides a hydrophilic protective layer around the nanoparticles which repel the adsorption of opsonin proteins via steric repulsion forces, thereby blocking and delaying the first step in the opsonisation process (37). PEGylated PLGA nanoparticles contains a hydrophilic coating of PEG and hydrophobic core of PLGA. An attempt was made to carry both Dalargin (hydrophilic drug) and Loperamide (hydrophobic drug) using PEGylated PLGA nanoparticles. Dalargin got adsorbed on the hydrophilic coat of PEG while Loperamide was entrapped in the hydrophobic core of PLGA. In vitro evaluation showed quick release of Dalargin as free drug while Loperamide HCl showed almost sustained release profile (38). Dalargin loaded PLGA nanoparticles was double coated with polysorbate 80 and polyethylene glycol (PEG). Polysorbate 80 coating provides protection against phagocytosis and PEG provides long circulating characteristics. The Dalargin-loaded polybutylcyano acrylate (PBCA)-nanoparticles were coated by adding up to 2% of Tween 80 and PEG 20000 stepwise to the nanoparticle suspension and kept under continuous magnetic stirring at 9000 rpm for 45 min (39).

PEGylated PLGA nanoparticles reported to carry Cytarabine to brain (40). Confocal microscopy evidenced the fluorescent PEGylated Cytarabine loaded PLGA nanoparticles in brain and spinal cord. It is reported that PEGylated polyhexadecylcyanoacrylate (PHDCA) nanospheres are good carrier for brain tumour targeting. Probable mechanisms include reduction of blood plasma clearance due to diffusion of nanoparticles across the brain, translocation due to the adsorption of PEGylated nanospheres to the brain endothelial cells (40). A PEGylated polymeric nanoparticle penetrates brain better than polysorbate 80 coated nanoparticles due to the fact that the covalent attachment of polyethylene glycol (PEG) to the polymer prevents desorption of PEG from PEGylated polymeric nanoparticles (41) unlike polysorbate 80 which is adsorbed to the polymer. Till now several drugs are successfully brain targeted by using coated nanotechnology as shown in Table 3.

Ligand nanotechnology: This approach is based on the covalent linkage of ligands to the polymers or the nanoparticles in order to promote receptor mediated endocytosis or transporter mediated transcytosis (49). The ligands can be transferrin, lipoprotein, insulin and thiamine but also synthetic or natural peptides can be used (50). The ligands are attached to the nanoparticles or polymer surface by two techniques

Covalent Chemical conjugation (51, 52 and 53): This is the most commonly established method of chemical conjugation where initially thiolation of ligand is done that is subsequently reacted with maleimide-functionalized drug or nanoparticle to form a stable thioether bond. Thiolated drug or vector can also be reacted with a free cysteine or reduced disulfide bond to yield a disulfide-bonded drug-nanoparticles conjugate. To further ensure functionality of the vector and protein, a chemical spacer (CH$_2$)$_5$NHCO or polyethylene glycol (PEG) moiety can be incorporated into the linkage to reduce steric hindrance.

Noncovalent Streptavidin/ Biotin linkages (51, 54, 55 and 56): The therapeutics can be monobiotinylated at lysine residues using N-hydroxysuccinimide (NHS) analogs of biotin, or alternatively, biotin can be attached using biotin hydrazide. The streptavidin can be coupled to the targeting vector via a thioether linkage. A BBB-targeted therapeutic can then be created simply by mixing the biotinylated therapeutic with the streptavidin-functionalized targeting vector. A PEG linkage can be also used.

Transferrin: Transferrin and insulin are reported to be used for the first time in ligand based nanotechnology. Transferrin undergoes receptor mediated endocytosis via transferrin receptors highly expressed on the brain capillary endothelial cells. Transferrin conjugated Polylactide – co-

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glycolide (PLGA) nanoparticles could successfully target Nevirapine to brain (52). But Transferrin use in ligand based nanotechnology is limited because blood plasma is almost saturated with endogenous Transferrin. The drug targeting Transferrin competes with the endogenous transferrin for the same transferrin receptor localised in brain endothelial cell, in turn it reduces the efficacy of transferrin conjugated nanoparticles as a carrier to deliver the desired therapeutic concentration of drug to brain. Hence antibodies are used in place of transferrin to overcome its limitation (53). One such antibody is ox26 which is reported to bind an extracellular epitope of transferrin distinct from transferrin binding site, and prevents competition between the drug targeting ligand and the natural endogenous ligand present in blood plasma. The ox26 is attached to the formulation by covalent chemical linkages, where thiolated ox26 antibody is conjugated to the maleimide-grafted liposomes according to a sulfhydryl-maleimide coupling method (54). One of the relevant work reported is the delivery of Tempol across BBB. Ox26 antibody covalently attached to maleimide grafted PLGA nanoparticles using NHSPEG 3500 maleimide crosslinker was a successful carrier for Tempol into brain (55). Attempts were made on the preparation of PEGylated immunonanoparticles (15). One such example is ox26 antibody conjugated PEGylated polyactic acid nanoparticles. Moreover polymers other than polyactic acid are also applied such as Chitosan. Chitosan- PEG nanospheres conjugated with ox26 were prepared by Avidin- Biotin complex. In this technique, Biotin was covalently coupled to PEG followed by covalent coupling of Chitosan to lead to a Chitosan- PEG Biotin copolymer. In

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<th>Drugs</th>
<th>Categories</th>
<th>Techniques of coated nanotechnology</th>
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<tbody>
<tr>
<td>Tacrine</td>
<td>Antialzheimer drug</td>
<td>Tacrine loaded polybutylcyanoacrylate nanoparticles coated with polysorbate 80.</td>
<td>(19)</td>
</tr>
<tr>
<td>Dalargin</td>
<td>Peptide</td>
<td>Polysorbate 80 coated Dalargin loaded PBCA nanoparticles</td>
<td>(42)</td>
</tr>
<tr>
<td>Donepezil</td>
<td>Antidementia drug</td>
<td>Donepezil loaded polybutylcyanoacrylate (PBCA) nanoparticles coated with polysorbate 80</td>
<td>(43)</td>
</tr>
<tr>
<td>Resperidone</td>
<td>Antipsychotic Drug</td>
<td>Poloxamer coated Resperidone loaded poly (epsilon-caprolactone) nanoparticles.</td>
<td>(44)</td>
</tr>
<tr>
<td>Amphotericin B</td>
<td>Antifungal drug</td>
<td>Amphotericin B loaded poly (lactic acid) – b- poly (ethyleneglycol) nanoparticles coated with polysorbate 80.</td>
<td>(45)</td>
</tr>
<tr>
<td>Resperidone</td>
<td>Antipsychotic Drug</td>
<td>Poloxamer 407 coated Resperidone loaded PLGA nanoparticles.</td>
<td>(46)</td>
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<tr>
<td>Estradiol</td>
<td>Hormones</td>
<td>Estradiol loaded polylactide–co-glycolide (PLGA) nanoparticles coated with polysorbate 80</td>
<td>(47)</td>
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<tr>
<td>Methotrexate</td>
<td>Antifungal Drug</td>
<td>Polysorbate 80 coated Methotrexate loaded chitosan and glycolchitosan nanoparticles.</td>
<td>(48)</td>
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</tbody>
</table>

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parallel, Streptavidin/ox26 conjugate is prepared and incubated with chitosan-PEG Biotin nanoparticles (prepared by ionotropic gelation technique using pentasodium tripophosphate as crosslinking agent) to obtain immunonanoparticles (56). PEGylated immunonanoparticles carried caspase inhibitor (peptide z DEWD-FMK) across BBB and reduced the death of neuronal cells after an ischaemic attack. It is also reported that transferrin conjugated PEGylated albumin immunonanoparticles could carry Azidothymidine significantly to brain as observed in rat (57).

**Insulin:** Insulin is not a suitable ligand based nanotechnology because of rapid degradation in blood stream (serum half-life 10 minutes) and hypoglycaemia due to possible interference with natural insulin balance (58). So, antibody recognising insulin receptors are used as brain targeting ligands. Researches using 83-14 mouse monoclonal antibodies (mAb) against insulin receptor for receptor mediated endocytosis were performed in primates (Rhesus monkey) (59). Attempts were also made to cure mucopolysaccharodosis type VII due to lysosomal deficiency. α-glucoridinase, an essential enzyme for lysosomal deficiency, was administered as radiolabelled phosphorylated glucoridinase (131I-P-GUS). Glucoridinase was found to act on mannose-6-phosphate receptor (Insulin like growth factor II) expressed on the endothelial cells of brain and gets delivered via receptor mediated endocytosis (59). Mannose-6-phosphate receptors in brain could be beneficial for ligand nanotechnology in order to treat many neurodegenerative disorders.

**Thiamine:** Thiamine (a water soluble vitamin B1), a micronutrient essential for normal cell growth and development is reported to cross the BBB by carrier mediated transport system (60). Thiamine as a surface ligand on the nanoparticles specifically targets them to the brain via the BBB thiamine transporter. Thiamine coated solid lipid nanoparticles comprising of emulsifying wax and Brij 78, were reported to act on thiamine transporter in brain as tested in situ by rat perfusion technique (61).

**Peptide derived nanoparticles:** Several peptide transport mechanisms (receptor mediated, adsorptive mediated, carrier mediated, nonspecific passive diffusion) as well as nontransport processes (endocytosis without transcytosis, absorption and metabolism) are reported. Several strategies are followed to manipulate peptide transport across the BBB so as to deliver drug to brain such as lipidization, chemical modifications of the N-terminal in peptides, coupling of transport with post BBB metabolism and formation of potent neuroactive peptides, upregulation of putative peptide transporters, use of chimeric peptides in which nontransportable peptide is chemically linked to a transportable peptide, use of monoclonal antibodies against peptide receptors and binding of circulating peptides to apolipoproteins (62). Researchers’ focuss on manipulating these strategies to target compounds/drugs to brain. One such reported successful work is on 12-32 (g21) of leptin conjugated PLGA nanoparticles which was successfully brain targeted as the confocal microscopy evidenced labelled tetramethylrhodamine g21 conjugated PLGA nanoparticles presence in rat brain (61). Another work is also reported on nanoliposomes containing phosphatidic acid or cardiolipin, which were decorated with two apolipoproteins (ApoE) derived peptides (the fragment 141-150 or its tandem dimers) for brain targeting. Confocal microscopy revealed enhanced brain uptake of nanoliposomes containing phosphatidic acid decorated with fragment 141-150 than its tandem dimers (63). It is reported that 29 amino acid peptide derived from rabies virus glycoprotein (RVG29) peptide conjugated to albumin nanoparticles using noncovalent streptavidin/biotin linkage significantly facilitate the intracellular delivery of nanoparticles as studied in vitro (64). One relevant work is reported, on viral fusion peptide (gH625) derived from the glycoprotein gH of Herpes Simplex virus type 1 covalently bound to the surface of fluorescent aminated polystyrene nanoparticles, which is found to be an efficient carrier for targeting therapeutics to brain. The gH625 covalently bound to polystyrene nanoparticles
Table 4. Drugs brain targeted through conjugated nanotechnology

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<th>Drugs</th>
<th>Categories</th>
<th>Techniques of coated nanotechnology</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ritonavir</td>
<td>Antiretroviral drugs</td>
<td>TAT conjugated Ritonavir loaded Polylactide(PLA) nanoparticles</td>
<td>(68)</td>
</tr>
<tr>
<td>Human serum</td>
<td>protein</td>
<td>HSA nanoparticles covalently bound with apolipoprotein.</td>
<td>(69)</td>
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<tr>
<td>albumin(HSA)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nevirapine</td>
<td>Anti retroviral drugs</td>
<td>Nevirapine loaded PLGA nanoparticles conjugated with transferrin.</td>
<td>(70)</td>
</tr>
<tr>
<td>Loperamide</td>
<td>Antinociceptive drug</td>
<td>Loperamide loaded HSA nanoparticles covalently coupled with insulin or antiinsulin receptor monoclonal antibody (29B4)</td>
<td>(71)</td>
</tr>
<tr>
<td>Coumarin 6</td>
<td>Anticoagulant</td>
<td>Coumarin 6 loaded PLGA nanoparticles conjugated with solanum tuberosum lectin.</td>
<td>(72)</td>
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<tr>
<td>Zidovudine</td>
<td>Antiretroviral drug</td>
<td>CRM 197 grafted Zidovudine loaded polybutyl cyanoacrylate (PBCA) nanoparticles.</td>
<td>(73)</td>
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Table 5. Drugs brain targeted through modification of polymers

<table>
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<th>Categories</th>
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<th>References</th>
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<tbody>
<tr>
<td>Didanosine</td>
<td>Antiviral drug</td>
<td>Didanosine loaded chitosan crosslinked with tripolyphosphonate anions nanoparticles.</td>
<td>(79)</td>
</tr>
<tr>
<td>Estradiol</td>
<td>Hormone</td>
<td>Estradiol loaded chitosan crosslinked with tripolyphosphonate anion nanoparticles.</td>
<td>(80)</td>
</tr>
<tr>
<td>Lamivudine</td>
<td>Antiretroviral drug</td>
<td>Lamivudine loaded chitosan crosslinked with glutaraldehyde nanoparticles.</td>
<td>(81)</td>
</tr>
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</table>

nanoparticles could be easily uptaken by brain as shown by endothelial cells BBB models. It is found that gH625 has high cell translocation potency; the peptide is free of toxicity, and also decreases nanoparticles intracellular accumulation (65). A significant work is reported on Chitosan conjugated pluronic based nanocarrier with a specific target peptide (rabies virus glycoprotein, RVG29) as a successful carrier for the delivery of protein (α galactosidase) to brain significantly (66). Cyclophillin B (Cyclosporin A binding protein) is reported to undergo receptor mediated transcytosis as observed in in vitro model of BBB. Cyclophillin B enables promoting regeneration of damaged peripheral nerves in addition to immunosuppressive activity (67). As cyclophillin B can cross BBB, so it may be utilised in peptide derived nanoparticles for treating brain related disorders. Several drugs efficiently delivered into brain using ligand nanotechnology are given in table 4.

**Nanotechnology based on modification of polymer:** Both synthetic and natural polymers can be used in nanotechnology for brain targeted drug delivery. One such interesting natural
<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Drugs</th>
<th>Technique of surface modification of nanoparticles</th>
<th>Blood brain barrier crossing ability of</th>
<th>References</th>
</tr>
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<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Surface modified nanoparticles</td>
<td>Nanoparticles without any surface modifications</td>
</tr>
<tr>
<td>1</td>
<td>Dextran</td>
<td>PLA nanoparticles coated with Tween 80.</td>
<td>It could cross the blood brain barrier as observed under florescence microscope</td>
<td>It could not cross the blood brain barrier.</td>
</tr>
<tr>
<td>2</td>
<td>Tacrine</td>
<td>Tween 80 coated polybutylcyanoacrylate (PBCA) nanoparticles loaded with Tacrine.</td>
<td>A modified nanoparticle has higher concentration of Tacrine in brain upon intravenous administration to rats.</td>
<td>A higher concentration of drug tacrine was observed in liver, spleen and lungs with the unmodified nanoparticles</td>
</tr>
<tr>
<td>3</td>
<td>Dalargin</td>
<td>Polycyanooacrylate (PBCA) nanoparticles double coated with Tween 80 and polyethylene glycol (PEG) 20000.</td>
<td>A central antinociceptive effect of Dalargin by tail flick test in mice is reported.</td>
<td>Absence of central antinociceptive effect of Dalargin by tail flick test in mice is reported</td>
</tr>
<tr>
<td>4</td>
<td>Amphotericin</td>
<td>Tween 80 coated polylactic acid-b-polyethylene glycol (PLA-b-PEG) nanoparticles.</td>
<td>The colony growth of cryptococcus neoformans in brain of mice is reduced as observe in vivo.</td>
<td>It could not reduce the growth of cryptococcus neoformans in brain of mice as observe in vivo.</td>
</tr>
<tr>
<td>5</td>
<td>Didanosine</td>
<td>Mannan coated Didanosine loaded Gelatin nanoparticles.</td>
<td>12.4 times higher uptake of Didanosine was found in brain than Didanosine administered in phosphate buffer solution intravenously.</td>
<td>It is not able to cross the blood brain barrier as reported by florescence study.</td>
</tr>
<tr>
<td>6</td>
<td>Ritonavir</td>
<td>Tat-conjugated ritonavir-loaded nanoparticles</td>
<td>The HIV infection of monocyte derived macrophages (MDM) cultures could be reduced.</td>
<td>The HIV infection of monocyte derived macrophages (MDM) could not be reduced.</td>
</tr>
<tr>
<td>7</td>
<td>Didanosine</td>
<td>Chitosan nanoparticles crosslinked with tripolyphosphate anions.</td>
<td>A significantly higher concentration (&lt; 0.5) of Didanosine is reported in brain after intranasal administration than that of intravenous administration.</td>
<td>Chitosan is fragile in nature. So, there is a chance of breakage of polymer if it is prepared without crosslinking.</td>
</tr>
<tr>
<td>8</td>
<td>Resperidone</td>
<td>Poloxamer coated poly (lactide co glycolide) nanoparticles.</td>
<td>A prolonged antipsychotic effect of Resperidone for 72 hrs was obtained upon subcutaneously administered to mice with lesser extrapyramidal side effects.</td>
<td>No antipsychotic effect is reported as uncoated nanoparticles could not cross blood brain barrier.</td>
</tr>
</tbody>
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Brain targeting nanoparticles
| 9  | Methotrexate | Tween 80 coated chitosan or glycol chitosan nanoparticles. | Tween 80 coated fluorescent chitosan nanoparticles transport Methotrexate across MDCKII-MDR1 cells | Uncoated nanoparticles could not transport Methotrexate across MDCKII-MDR1 cells | 48 |
| 10 | Human serum albumin (HSA) | Tween 80 coated or covalently bound apolipoprotein E (Apo E) HSA nanoparticles | HSA is reported to be transferred across brain capillary endothelial cells and neurons when injected intravenously into SV 129 mice under transmission electron microscopy | Unmodified surface of nanoparticles unable to cross the brain capillary endothelial cells and neurons when injected intravenously into SV 129 mice under transmission electron microscopy | 69 |
| 11 | Nevirapine | Transferrin-grafted poly (lactide-co-glycolide) nanoparticles loaded with Nevirapine. | Diododecyldimethylammonium bromide (DODAB)-stabilized Nevirapine loaded poly (lactide-co-glycolide) nanoparticles grafted with Transferrin enhances the transport of Nevirapine (NVP) across human brain microvascular endothelial cells (HBMECs) | Unmodified surface of Nevirapine loaded poly (lactide-co-glycolide) nanoparticles is not reported to cross human brain microvascular endothelial cells (HBMECs) | 70 |
| 12 | Loperamide | Insulin or an anti-insulin receptor monoclonal antibody (29B4) covalently coupled Loperamide loaded Human serum albumin(HSA) | Induction of antinociceptive effects in the tail-flick test in ICR (CD-1) mice after intravenous injection | Noninduction of antinociceptive effects in the tail-flick test in ICR (CD-1) mice after intravenous injection | 71 |
| 12 | Coumarin 6 | Solanum tuberosum lectin (STL) conjugated poly (DL-lactic-co-glycolic acid) (PLGA) nanoparticle (STL-NP). | Solanum tuberosum lectin (STL) conjugated poly (DL-lactic-co-glycolic acid) (PLGA) nanoparticle (STL-NP) demonstrated 1.89-2.45 times (p < 0.01) higher brain targeting efficiency than unmodified NP of Calu-3 cells. Enhanced accumulation of STL-NP in the brain is reported by near infrared fluorescence probe image following intranasal administration | Unmodified nanoparticles lesser brain targeting efficiency than modified nanoparticles. | 72 |
| 13 | Zidovudine | CRM197-grafted polybutylcyanoacrylate (PBCA) nanoparticles loaded with Zidovudine. | Modified nanoparticles could transverse monolayer of human brain-microvascular endothelial cells (HBMECs) | Unmodified nanoparticles could not transverse monolayer of human brain microvascular endothelial cells (HBMECs) | 73 |
### Table 7. Patents for nanoparticle based CNS targeted drug delivery systems

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Application</th>
<th>Summary of invention</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Receptor targeted drug delivery systems</td>
<td>Chemical conjugate of polymeric nanoparticles for brain targeted delivery</td>
<td>86</td>
</tr>
<tr>
<td>2</td>
<td>Drug targeting system, method for preparing same and its use</td>
<td>Dalargin loaded nanoparticles coated with polysorbate 80</td>
<td>87</td>
</tr>
<tr>
<td>3</td>
<td>Transport of liposomes across the blood-brain barrier</td>
<td>Monoclonal antibodies (mAb) conjugated liposomes for brain targeted delivery.</td>
<td>88</td>
</tr>
<tr>
<td>4</td>
<td>Drug targeting system, method of its preparation and its use</td>
<td>Dextran 12.000 or polysorbate 85 stabilized nanoparticles for brain targeted delivery of Dalargin.</td>
<td>89</td>
</tr>
<tr>
<td>5</td>
<td>Use of drug loaded nanoparticles for the treatment of cancers</td>
<td>Coated nanoparticles for the delivery of anticancerous drug (Doxorubicin) to brain.</td>
<td>90</td>
</tr>
<tr>
<td>6</td>
<td>Non-invasive gene targeting to the brain</td>
<td>Ox26 monoclonal antibodies conjugated polyethylene glycol (PEG) immunoliposomes for brain targeted delivery.</td>
<td>91</td>
</tr>
<tr>
<td>7</td>
<td>Nanoparticles made of protein with coupled apolipoprotein E for penetration of the blood-brain</td>
<td>Avidin-modified Human serum albumin (HSA) nanoparticles with biotinylated apoE for brain targeted delivery.</td>
<td>92</td>
</tr>
<tr>
<td>8</td>
<td>Non-invasive gene targeting to the brain</td>
<td>OX26 MAb conjugated PEGylated liposome for gene delivery</td>
<td>93</td>
</tr>
<tr>
<td>9</td>
<td>Support system in the form of protein-based nanoparticles for the cell-specific enrichment of pharmaceutically active substances</td>
<td>Preparation of nanoparticles by miniemulsion; surface modification by coating with polysorbate 80 for brain targeting.</td>
<td>94</td>
</tr>
<tr>
<td>10</td>
<td>Rapid Diffusion of Large Polymeric Nanoparticles in the Mammalian Brain</td>
<td>Polyethylene glycol (PEG) coated polymeric nanoparticles loaded with drug and gene for brain targeted delivery.</td>
<td>95</td>
</tr>
<tr>
<td>11</td>
<td>Drug delivery in neurodegenerative disorders</td>
<td>Nanoparticles loaded with epidermal growthfactor</td>
<td>96</td>
</tr>
<tr>
<td>12</td>
<td>Nanoparticles for protein drug delivery</td>
<td>Nanoparticles composed of chitosan and polyglutamic acids for the brain targeted delivery of protein or bioactive agents.</td>
<td>97</td>
</tr>
</tbody>
</table>
13 Encapsulation of biologically active agents

Encapsulation of biologically active agents such as proteins in particulate carriers such as nanoparticles using Hydrophobic ion pairing (Hip) agents

98

14 Polylactide nanoparticles

Pluronic 188 coated drug loaded poly (lactide co glycolide) nanoparticles for brain targeting.

99

15 Nanoparticles made of protein with coupled apolipoprotein e for penetration of the blood-brain barrier and methods for the production thereof

Human serum albumin (HSA) avidin nanoparticles conjugated with ApoE for brain targeted delivery of Dalargin.

100

16 Conjugates for targeted drug delivery across the blood-brain barrier

Conjugates of Distearoylphosphatidyl ethanolamine-polyethylene glycol-maleimide (DSPE-PEG-MAL) with reduced glutathione was prepared for brain targeted delivery.

101

17 Rapid Diffusion of Large Polymeric Nanoparticles in the Mammalian Brain

Polyethylene glycol (PEG) coated polymeric nanoparticles loaded with drug and gene for brain targeted delivery.

101

18 Targeting of drugs and diagnostic agents

Conventional nanoparticles coated with surfactants to cross blood brain barrier

102

19 Site specific drug delivery across Blood brain barrier

Nanogels prepared from cross-linked polyon polymer fragment and one nonionic water soluble polymer fragment

102

20 Protein and peptide delivery to brain

Nanoparticles prepared from chitosan and polyglutamic acid

102

21 Inhibition of reperfusion injury to brain

Nanoparticles prepared from inert plasticizers loaded with anti-oxidants

102

Table 8. FDA approved CNS targeted drug delivery systems using nanoparticles (102)

<table>
<thead>
<tr>
<th>Sr. No</th>
<th>API/ nanoparticle components</th>
<th>Route of administration</th>
<th>FDA approved indication</th>
<th>Product</th>
<th>Company</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Propofol</td>
<td>Intravenous</td>
<td>Anesthetic</td>
<td>Diprivan</td>
<td>Zenechpharma</td>
</tr>
<tr>
<td>2</td>
<td>Colloidal gold nanoparticles coupled to TNF and PEG-Thiol (~27 nm)</td>
<td>Intravenous</td>
<td>Solid tumors</td>
<td>Aurimmune (CYT-6091)</td>
<td>Cytimmune Sciences</td>
</tr>
<tr>
<td>3</td>
<td>Cyclodextrin containing siRNA Delivery nanoparticles (~50 nm) based on Calando’s RONDEL technology</td>
<td>Intravenous</td>
<td>Cancer</td>
<td>CALAA-01</td>
<td>Calando Pharmaceutical</td>
</tr>
<tr>
<td>4</td>
<td>Gold-coated silica nanoparticles (~150 nm)</td>
<td>Intravenous</td>
<td>Solid tumors</td>
<td>AuroShell</td>
<td>Nanospectra Biosciences</td>
</tr>
</tbody>
</table>

Sunita Lahkar and Malay K Das
polymer extensively used in the nanotechnology field due to its nanoparticles forming ability is Chitosan. Chitosan has several characteristics favouring its use in preparing brain targeted nanoparticles such as it is natural, biodegradable, biocompatible, bioadhesive, low molecular weight (LMW) (74). Inspite of these advantages, chitosan nanoparticles suffer from fragile structure, making it unsuitable to use without modification as carrier for drug molecules. Several techniques of modifications are suggested, but the simpler technique (called Ionotropic Gelation) is through chitosan salt formation where some anions may cause crosslinking via ionic interactions (75). In this connections, tripolyphosphonate, sodium citrate, amino acids, sodium sulphate can be used as crosslinkers (76-78, 74). The drugs delivered across the BBB using this technology are given in Table 5.

Conclusion

The Blood brain barrier (BBB) is the most limiting condition for the efficient drug delivery to CNS. Nanoparticles have good prospect in treating brain disorders. It has major contribution in the delivery of inaccessible drug to the brain and thus also helps in treating brain cancer or other neurodegenerative disorder. The pharmacokinetics, patented technology and FDA approved CNS targeted nanoparticles with different drugs are shown in Table 6-8, respectively. In near future nanoparticulate drug delivery systems can be used for exploiting many biological drugs which have poor aqueous solubility, permeability and less bioavailability. Nanoparticles provide ingenious treatment of CNS disorders by enabling targeted delivery and controlled release. Thus nanoparticles can be considered to be significant in brain targeting drug delivery. Nanoparticulate drug delivery technology should be developed further which can be achieved by prompt participation of more research oriented programmes from the governmental as well as corporate sectors.

References


Brain targeting nanoparticles


63. Re, F., Cambianica, I., Sesana, S., Salvati, E., Cagnotto, A., Salmina, M., Couraud, P.O.,


