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Exploring the therapeutic potential of neuropeptides in neurodegenerative disease (NDD): A review

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Abstract

Neurodegenerative diseases (NDD), in which neuronal cells disintegrate, bring about deteriorations in cognitive functions as is evidenced in millions of patients. The use of therapeutic potential of neuropeptides (NP's) or treating CNS disorders has attracted the interest of the scientific community. NP's are expressed and released by neurons, and mediate neuronal communication by acting on cell surface receptors and are presented as the processed, biologically active products of the neuropeptide precursors alongwith the encoding gene. As CNS (Central nervous system) is one of the most eminent part of the brain which drives very crucial sensory motor activities in human body. Moreover, any physiological restrictions like BBB (blood brain barrier), CSF (cerebro spinal fluid) etc. may lead to non transportation of drug molecules in diseased conditions, specifically NDD's. Therefore, the role of neuropeptides for the treatment of same is been explored heavily as they are able to pass through this barrier without being altered or denatured and have proved to possess significant therapeutic potential against NDD's. The present paper focuses on structure and functions of various neuropeptides like Nerve growth factor (NGF), Activity derived neurotrophic factor (ADNF) and Vasoactive intestinal protein (VIP) etc. for their properties by which they help in treating neurodegeneration process. Further, different strategies and formulations of targeted drug delivery of neuropeptides like addition of NGF loaded microspheres to PC12 cell line, intranasal administration of fatty neuropeptide and nanoparticle for growth factor delivery were also discussed. These observations on the carriage and release of growth factors by the proposed mechanisms open new therapeutic options for both neuronal regeneration and for the development of effective neuronal interfaces.

Keywords

Neuropeptide, Neurodegeneration, Targeted drug delivery, NGF, Magnetic micro particles, Neuronal outgrowth.

Introduction

Neurodegeneration is a process which denotes the delayed progressive loss of structure or function of neurons at many different levels of neuronal electrical system ranging from molecular to systemic. Degenerative nerve diseases cause worsening of many activities, including balance, movement, talking, breathing and heart functions. Certain examples of degenerative nerve diseases (Gozes and Brenneman, 1999) includes- Alzheimer's disease (AD), Huntington's disease (HD), Parkinson's disease (PD), Spinal muscular atrophy (SMA). The treatment initiatives usually involve, Improvement in symptoms, relieve pain and increase mobility. The greatest risk factor for NDD's is aging and mitochondrial DNA mutations as well as oxidative stress both contribute to the process of aging. Also, protein degradation offers therapeutic options both in preventing the synthesis and degradation of irregular proteins. There is also interest in up regulating autophagy to help clear protein aggregates implicated in neurodegeneration (Gozes and Brenneman, 1999). Here, we are discussing the role of therapeutic neuropeptides to cure neurodegenerative diseases.

Neuropeptides are small chains of amino acids used by neurons to communicate signals with each other. They are neuro signaling molecules that influence the activity of signal transferring in the brain. NP's are expressed and released by neurons, and modulate neuronal communication by acting on cell surface receptors. Many NP's are co-released with other small-molecule neurotransmitters. The human genome contains about 90 genes that encode precursors of neuropeptides. At present about 100 different peptides are known to be released by different populations of neurons in the mammalian brain.

Functional mechanism of NP's

Generally, peptides act on G-protein -coupled receptors (GPCR) expressed by selective populations of neurons. In essence they act as specific signaling molecules between one population of neurons with another one whereas; neurotransmitters generally affect the excitability of neurons. NP's have much more diverse effects like they can affect gene expression, local blood flow, synaptogenesis, and glial cell morphology. Moreover, they tend to have prolonged actions, and some of them have striking effects on behavior too (Wiesmanna and De Vos, 2000). Neurons very often make both a conventional neurotransmitter (such as glutamate or dopamine) and one or more neuropeptides.

Properties of neuropeptides

They are natural so unlike, other conventional drugs that undergo hepatic metabolism and cause side effects in the body, they show no such kind of activity. NP's usually increases the efficiency of synaptic activity more than conventional neurotransmitters and hence, are projected as suitable therapeutic targets against many NDD's. But, although being effective therapeutic strategy, they have a structural limitation as they get degraded easily, when the surrounding temp or pH changes. This property of NP's is helpful in a way as it avoids side effects alongwith accumulation but it creates a problem of persistence (Steingart, 2000). So, to improve upon, on this restriction various formulation approaches and delivery routes are been explored.

Biological action of NP's in CNS

The biological action of neuropeptides significantly in CNS area is dependent on their type as there are some specific NP's like - VIP (Vasoactive Intestinal Peptide), ADNF (Activity-Dependent Neuroprotective Factor), NGF (Nerve Growth Factor) which have designated relevant activities for initiating and maintaining neuronal mechanism. Alzheimer's disease and Parkinson's disease are two neurodegenerative disorders which are being researched upon

intensively. After rigorous researches it has been found that NGF (Nerve Growth factor), VIP (Vasoactive Intestinal Peptide), ADNF (Activity Dependent Neurotrophic Factor) and Neuropeptide Y are among those neuropeptides which have shown great activity against these diseases.

VIP is one such peptide hormone belonging to the glucagons family that binds to a member of the class II family of G protein-coupled receptors (GPCRs) – VPAC1, VPAC2, and PAC1. It's mechanism is activated by the binding to VCAP1 and VCAP2 receptors present on astrocytes which further leads to the initiation of PKC (Protein Kinase C) mechanism, a major component of astrocyte survival (Delgado and Ganea, 2013). Once the binding has taken place successfully then the whole mechanism begins. This binding stimulates AC (adenylate cyclase) which increases intracellular cAMP leading to increase in the production of Ca^{+} intracellularly. This further increases nuclear translocation of protein kinase C (PKC) (Khan and Tiwari, 2012) thus, promoting astrocytic survival. Mechanism of VIP action promotes release of various astroglia derived factors that provide neuroprotection such as:-

- Interleukin1 α released, is a neuroprotective factor regulated in astrocytes by VIP (Brenneman et al., 1997).
- VIP releases proteins from astrocytes which are protease inhibitor, Protein Nexin 1.

Similarly, VIP is another NP's that releases complex array of cytokines from astrocytes that contributes to the mitogenic and neurotrophic properties of this neuropeptide in CNS (Delgado and Ganea, 2013). Most importantly it releases ADNF (Activity Dependent Neurotrophic Factor) on binding to astrocytes which is a major neuroprotection factor. This neuroprotection is also carried out by NAP (Asn- Ala- Pro- Val- Ser- IIe-Pro-Gln) (Gozes et al., 2003). After being released from astrocytes, ADNF binds to G protein coupled receptors on neural membrane. After binding to its receptors it starts a cascade of events (particularly PKC and MAPK pathway) involving secondary messengers and other enzymes responsible for activation of proteins which help in DNA replication and transcription (Lodish, 2000).

G proteins in contact with GPCRs are activated which further activate Phospholipase D. PLC helps in generating Diacyl glycerol (DAG) and Inositol triphosphate which cause intracellular mobilization of calcium ions which further activate protein kinase C (Kurakhmaeva et al., 2009) calcium and PKC activation phosphorylate Tyrosine kinase (TK) which in turn activates Ras proteins which form dimer with Raf-1 eventually leading to activation of MAPK (Mitogen Activated Protein Kinase) pathway. Activation of MAPK enhances the feedback loop. Both PKC and MAPK pathway are responsible for cell growth (Hang and Liu, 2002). MAPK pathway particularly regulates the activity of transcription factors and alters the transcription of genes required for cell cycle (Whitmarsh, 2007). Ultimately these pathways lead to neuroprotection. Some of the most important biological activities of ADNF are - It enhances microtubule formation and also helps in conduction of electrical impulses apart from preventing excitotoxicity. It plays an active role against oxidative stress and promotes neurite outgrowth. Also, it helps in synapse formation, blockage of electrical conductivity, excitotoxicity, beta amyloid peptide and oxidative stress. Now, any ligand binding to the GPCR II family has a great potential in treatment of CNS disorders. VIP and ADNF are those ligands and hence have immense potential in being used as a therapeutic neuropeptides (Gonzalez et al., 2010)

VIP is mainly used in treatment of Alzheimer's, spectrum disorders of autism, and Parkinson's. ADNF also finds therapeutic applications in treatment of Alzheimer's disease and other neurodegenerative disorders. In Alzheimer's, beta amyloid peptide aggregates into oligomers along microtubule and causes its depolymerisation (Mokhtar et al., 2013). ADNF enhances microtubule polymerization, prevents formation of reactive oxygen species (ROS) and prevents excitotoxicity. Studies have provided evidence that NGF is implicated in neurobehavioral response including cerebral alterations associated with psychiatric disorders. NGF (nerve growth factor) is a neurotrophin which is required in the development and maintenance of the peripheral and central nervous system. These highly homologous, homodimeric growth factors

control cell survival, differentiation, growth cessation, and apoptosis of sensory neurons (Verge et al., 1995). The biological functions of the neurotrophin are mediated through two classes of cell surface receptors, the Trk receptors and the p75 neurotrophin receptor ($p75^{NTR}$). Nerve growth factor (NGF), the best characterized member of the neurotrophin family, sends its survival signals through activation of Trk A and can induce cell death by binding to $p75^{NTR}$. NGF is synthesized and released within the central nervous system and exerts a trophic and functional role on basal forebrain cholinergic neurons; it is involved in a protective role following brain insults induced by an epileptic status, seizure, as well as surgical and chemical lesions. More recently in collaborative studies provided evidence that NGF is implicated in neurobehavioral response including cerebral alterations associated with psychiatric disorders (Egleton and Davis, 2005).

Possible role in treatment of neurodegenerative disorders

Neuropeptides in general and VIP in particular have been associated with normal brain development and have been attributed to having growth and survival promoting properties as well as influence on cognitive functions (Passemard et al., 2011). Many clinical trials are being conducted for VIP on various CNS disorders. Researches for ADNF are still under pipeline. Because of its maximum activity at femto molar concentrations it has come up as most sought after neuropeptide in the field of further research for treatment of neurodegenerative disorders.

For more than 35 years, NGF has been considered a very powerful and selective growth factor in sympathetic and sensory neurons and in cells deriving from the neuronal crest (Aloe et al., 2012). NGF is particularly involved in the functioning of the immunohematopoietic system and in physiological situations caused by neuroendocrine changes.

Recently, NGF has also been studied for its clinical application in peripheral neuropathies as induced by diabetes, leprosy, surgical traumas and in clinical trials on AIDS, as mentioned in a recent conference on NGF in relation to pathologies of the CNS as Alzheimer's and Parkinson's diseases (Bersani et al., 2000). One of clinical studies was recently published in the New England Journal of Medicine: for the first time the NGF therapeutic role is demonstrated in eye pathology as a neurotrophic keratitic ulcer, application of NGF rapidly recovers the lesion.

Current researches

In recent years, complex as well as contradictory results from experimental research have suggested that the factors which are active during the ontogenesis of the CNS may play a crucial role in the ethiopathogenesis of certain CNS related disorders like schizophrenia (Bersani et al., 2000). A great number of evidences published over the last few years suggest that the onset of schizophrenia in adulthood may be the consequence of early alterations in neurodevelopment encephalopathy. A possible role played by NGF in early alterations of the weak schizophrenic brain is also supported by the critical role played by neurotrophin during the primary development of cholinergic neurons which are implicated in memory and learning processes.
Neuropeptides in general and VIP in particular have been associated with normal brain development and have been attributed to having growth and survival promoting properties as well as influence on cognitive functions. Many clinical trials are being conducted for VIP on various CNS disorders. Researches for ADNF are still under pipeline. Because of its maximum activity at femtomolar concentrations it has come up as most sought after neuropeptide in the field of further research for treatment of neurodegenerative disorders. For more than 35 years, NGF has been considered a very powerful and selective growth factor in sympathetic and sensory neurons and in cells deriving from the neuronal crest. NGF is particularly involved in the functioning of the immunohematopoietic system and in physiological situations caused by neuroendocrine changes.

Moreover, brain is tightly segregated from the circulating blood by BBB, it represents a formidable obstacle for a large number of drugs, including the majority of anticancer agents, peptides and nucleic acids. As a consequence, this barrier prevents effective treatment of many severe and life threatening diseases like brain cancer and neurodegeneration (Upadhyay, 2004). The BBB is highly efficient and makes the brain practically inaccessible to lipid insoluble compounds. Brain delivery of such compounds therefore, requires a strategy to overcome the BBB. Advances in understanding of the cell biology of the BBB have opened new avenues and possibilities for improved drug delivery to CNS (Begley, 2003).

Another study related with VIP showed that, being a lipophilic analogue was prepared which enters the brain intact and testing has proven that it is A VIP analogue has greater potency and efficacy than VIP (Campos-Salinas J et al., 2014). VIP protects against Alzheimer-related retardation of learning and memory. It was seen that when oral dosage was given to mice the concentration entering the brain was negligible to carry out neuroprotection so new path was chosen for drug delivery which is intranasal administration. Intranasal administration showed positive results and it showed significant amount of concentration of the VIP analogue reaching the brain in intact form. [21]

Delivery pathways for neuropeptides:

Brain is tightly segregated from the circulating blood by a unique membranous barrier - the Blood Brain Barrier (BBB). The BBB represents a formidable obstacle for a large number of drugs, including the majority of anticancer agents, peptides and nucleic acids. As a consequence, this barrier prevents effective treatment of many severe and life threatening diseases like brain cancer and neurodegeneration (Prokop and Davidson, 2008). The BBB is highly efficient and makes the brain practically inaccessible to lipid insoluble compounds. Brain delivery of such compounds therefore requires a strategy to overcome the BBB. Advances in understanding of the cell biology of the BBB have opened new avenues and possibilities for improved drug delivery to the Central Nervous System (CNS).

Various strategies like non-invasive methods, including drug manipulation encompassing transformation into lipophilic analogues, prodrugs, chemical drug delivery, carrier-mediated drug delivery, receptor/vector mediated drug delivery and intranasal drug delivery, which exploits the olfactory and trigeminal neuronal pathways to deliver drugs to the brain, are widely used (Pathan et al., 2009). The lipophilic precursor scale, a possible choice for CNS prodrugs is to link the drug to a lipid moiety, such as a fatty acid, a glyceride or a phospholipid. Such prodrug approaches were explored for a variety of acid containing drugs, like levodopa. Problems associated with prodrugs are: the poor selectivity and poor tissue retention of some of these molecules. Besides, the lipidization strategy involves the addition of lipid-like molecules through modification of the hydrophilic moieties in the drug structure. Lipid-soluble molecules are believed to be transported through the BBB by passive diffusion but the lipid property of molecules generally increases the volume of distribution, particularly due to plasma protein-binding which affects all other pharmacokinetic parameters. Moreover, increasing lipophilicity tends to increase the rate of oxidative metabolism by cytochrome P-450 and other enzymes. While increased lipophilicity may improve diffusion across the BBB, it also tends to increase uptake into other tissues, causing an increased tissue burden. With all the formulation what finally is required is a route which has least number of barriers or pH, temp or surrounding conditions being changed, so that the peptide undergoes least number stress conditions and chances of its denaturation reduce (Upadhyay, 2014). By performing different tests scientists were able to come to a conclusion that intranasal administration proved to be a very good route so as to get neuropeptide concentration into the brain.

Formulation of Neuropeptides as Drugs for CNS Disorders

In recent years, there have been several important advancements in the development of neuropeptide therapeutics. The targeting of peptide drugs to the CNS remains a formidable obstacle. Delivery of peptide drugs is limited by their poor bioavailability to the brain due to low metabolic stability, high clearance by the liver, and the presence of the blood brain barrier (BBB). Multiple strategies have been devised in an attempt to improve peptide drug delivery to the brain, with variable results. In this review, we have discussed several strategies that have been used to improve both bioavailability and BBB transport. Further development of these delivery methods may finally enable peptide drugs to be useful for the treatment of neurological disease states (Egleton and Davis, 2005).
Neuropeptides have been indicated as primary molecules in several neurological disorders including epilepsy and depression. The use of peptides as pharmacological agents is an attractive proposition due to low toxicity of their metabolites and enhanced potency. Despite the growth in understanding of neurological disorders, peptide-based therapeutics are not currently available for treating these clinical problems. This is largely due to inadequate delivery of intact/viable peptides to specific brain regions necessary for neurological disease treatment. The delivery of peptide-based drugs to the brain is limited by two main factors; general bioavailability issues and the presence of the blood brain barrier (BBB). Various kinds of formulations used for neuropeptides are- PLGA microspheres, nanoparticulates, Liposomes, PEGylation and Hyperglycosilation

PLGA Microspheres and Nanoparticulate Drug Delivery

Polymeric nanoparticulates have been explored as drug delivery vehicles for decades. Poly (D, L-lactic-co glycolic-acid) (PLGA) microspheres and polybutylcyanoacrylate (PBCA) nanoparticles are the most successful formulation because of their high targeted drug delivery. PLGA microspheres for protein delivery can be prepared by techniques like spray drying, double emulsion and phase sepration-coacervation. The advantages are - biocompatible, biodegradable, increase release rates, high tensile strength, thermo plasticity, crystallinity. It is easily administered through injections and balances the negative effects on protein stability during preparation and storage. But it also has certain limitations like degradation which leads to accumulation of lactic and glycolic acids within drug delivery device resulting in reduction in pH of the micro-environment leading to denaturation of encapsulated protein. Harmful reactions may occur such as deamidation and theol catalysed di sulphide exchange (Barry and Vertegel, 2013). The product in the market include PBCA-NP (polybutylcyanoacrylate nanoparticulate)

PEGylation

PEGylation provided the real breakthrough in enhancing the pharmaceutical properties of protein and peptides in a viable manner PEGylation, the covalent attachment of PEG moieties to a therapeutic agent, was firstly reported in the 1970s. PEGylation results in overall enhancement of stability, pharmacokinetics and therapeutic utility of molecules.
PEG moieties are repeating units of ethylene glycol that are inert and amphiphillic. PEG is made from anionic ring opening polymerization of ethylene oxide. PEG molecules are linear or branched (Pisal et al., 2010). This process of PEGylation results in the formation of stable covalent bond between PEG polymers and drug. PEG is first activated by preparing functional groups at terminal end for which the most common choices are lysine and proteins N-terminal amino group. Carboxylic acid intermediates of PEG allowed 97% removal of impurities and incorporation of degradable linkages helped in the release of drugs in the cell. Site specific PEGylation can be done by reductive alkylation with PEG aldehyde. Example of an enzyme of the same is trans-glutaminase. These are biodegradable in nature and regarded as non-immunogenic. Disadvantages include PEG may build up over the tissues as a result of chronic

administration of agents with extensive distribution and is not better that hyperglycosilated products. The product in the market includes ONCASPAR and ADAGEN.

Hyperglycosilation

Glycosilation has been the most heavily studied posttranslational peptide modification. The carbohydrates attached to the protein help in determining the structure, function, activity, immunogenicity and pharmokinetics. For recombinant proteins this technique is highly dependent on the cell while the machinery necessary for glycosilation is absent from bacterial expression system (Pisal et al., 2010). Hyperglycosilation resembles the technique of PEGylation. PEG is an exogenous and does not get degraded easily but glycol groups are easily broken down. Example is polysialic acids (PSA). These can extend biological half-life, reduce immunogenicity and improve solubility. The glyco products are easily bio-degradable. Disadvantages are that the hyperglycosylated forms are expected to perform equally or reduced to their native counterparts. Product in market is Darbepoetin alpha, hyperglycosilatedform of EPO.

Possible role in treatment of neurodegenerative disorders

Peptides and proteins have become the most sought after drugs for the treatment of neurodegenerative diseases owing to their selectivity and their ability to provide a potent action. When the peptide drugs enter the human body they have to surpass many barriers for effective treatment of any neurological disease (Upadhyay, 2014). Their integrity has to be maintained and at the same time they should be able to reach the target site. One of the commonly adopted methods for peptide and protein drug administration is the oral route. It is the most preferred route because of its non-invasiveness and easier administration. When a protein enters the gastrointestinal tract (GIT), it is subjected to variations in pH which can degrade the protein molecule because of which they lose their activity (Khan et al., 2011). Secondly, the stomach is rich in proteolytic enzymes which act on the peptide bonds in the protein and break them into small oligopeptides. Most of the protein molecules are hydrophilic in nature. So even if some molecules are prevented from enzymatic degradation, they cannot pass through intestinal membrane because of their hydrophilic nature. Even if some of the molecules can pass through gut and enter the blood through portal circulation, they are transported to liver on their way into main blood pool. Liver has such enzymes which can easily cause breakdown of proteins and peptides. Hence proteins and peptides are unable to reach the main blood circulation in their fundamental form (Misra et al., 2003). Very small quantities of molecules which are able to cross intestinal membrane and enter the main blood circulation further encounter Blood Brain Barrier (BBB). The blood capillaries in brain are lined by endothelial cells which lack fenestrations and are sealed with tight junctions. So any molecule must pass through endothelial cell to reach the target cells in brain. The cell membrane is made up of lipid molecules, which allows easy penetration of lipid soluble substances through the cells. So, most of the proteins and peptides which are lipid insoluble and have high molecular weight are rendered useless as they cannot pass through BBB. To bypass these barriers novel methods of peptide administration are being developed. In order to increase the bioavailability of drugs various methods can be employed: modification of the chemical properties of the peptides i.e. manipulation of drugs, covalent attachment of certain compounds which modify the protein structure, disruption of BBB , use of improved delivery carriers and finding alternative routes for drug delivery (Shaji and Patole, 2008).

These drug delivery strategies can be classified as Invasive and Non-invasive approaches (Sharma et al., 2011). Different methodologies fall under these categories which are discussed briefly (Figure 1).

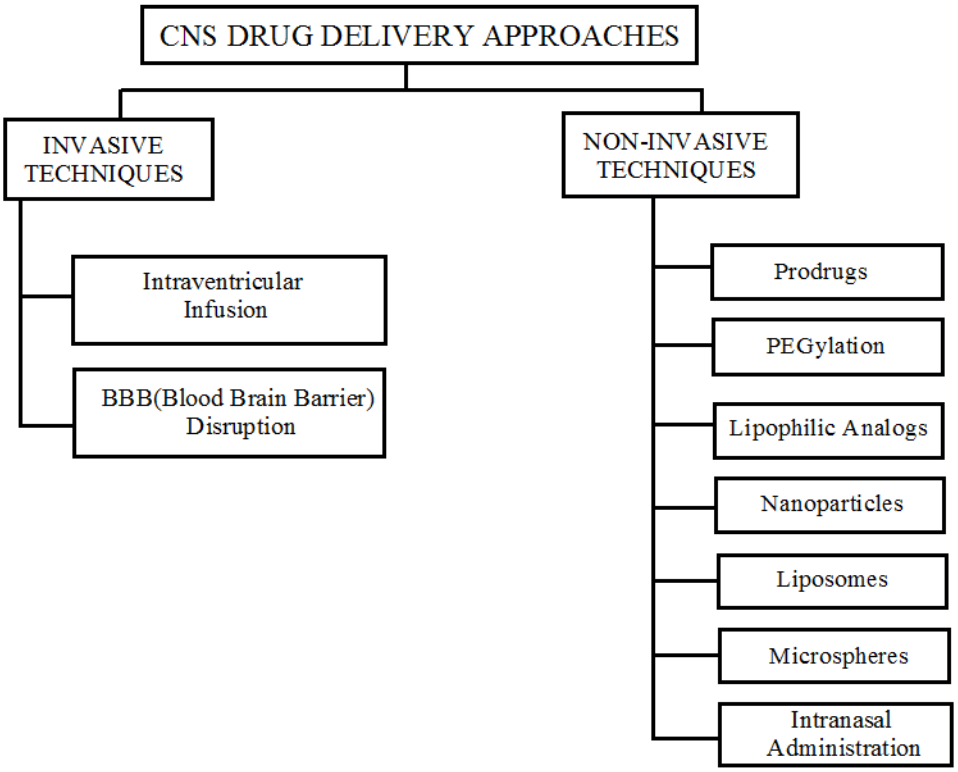

Figure 1: Image showing different strategies for CNS drug delivery

Invasive techniques are those strategies which cause a puncture or incision in the body. Two of the most commonly adopted methods of invasive techniques are Intra-ventricular infusion and disruption of BBB.
Intraventricular infusion: In this method a plastic reservoir (for example OMMAYA reservoir) is implanted subcutaneously in the brain and connected to the ventricles (cavities) in the brain. This leads to direct administration of drug in the cerebrospinal fluid (CSF). Here, drug reaches the target cells in very short period of time and it does not have to bypass the BBB. Also, drugs in CSF encounter minimized enzymatic degradation relative to drugs in plasma leading to a longer half-life.
Osmotic BBB disruption: In this method inert hypertonic solutions are injected in the veins which cause shrinkage of endothelial cells and opening of tight junctions of the BBB for a few hours. But this method has its own drawbacks like certain chemicals or toxins circulating in the blood can also cross this barrier thus leveling brain vulnerable to damage.

Non-invasive techniques are those in which drug properties are changed and different routes of administration are employed (Dwibhashyam and Nagappa, 2008).

a) Prodrugs: Biologically active compounds are chemically modified to form inactive compounds. Chemical changes are such that they improve some physicochemical property. For example, increasing membrane permeability and water solubility. When the prodrug reaches near the receptor site it is converted to active form and is maintained there for longer period of time. Generally hydroxyl, amino or carboxyl group containing drugs are modified by esterification or amidation. This enhances lipid solubility of the drug and hence easy entry into the brain.

b) PEGylation: PEG comprises of repeating units of ethylene glycol. These are both inert and amphiphilic. In this process stable covalent bonds are formed between PEG polymers and the peptide drug (Egleton and Davis, 2005). PEGylation increases the size and molecular weight of drugs. Increase in the radius of the molecule improves the

solubility in the blood and decreases the rate of filtration from glomerulus. So, PEGylated proteins show reduction in urinary clearance. Protective shell formed by PEGylating the peptides decreases the formation of antibodies against the proteins by masking the antigenic sites on their surface (Pisal et al., 2010). The process also increases hydrophilicity of the molecule so that it is not recognized by reticuloendothelial cells (for example macrophages) in the blood thus preventing their phagocytosis. It also stabilizes the molecules sterically resulting in circulation of molecules for longer time (Immordino et al., 2006).

c) Lipophilic analogues: Low molecular weight and lipophilic substances can easily pass through cell membranes (here membrane of endothelial cells). Most of the peptides used for treatment of neurodegenerative disorders are hydrophilic. These hydrophilic peptides are made hydrophobic or lipophilic by attaching lipid molecules to them (Mikitsh et al., 2014). It helps in easy penetration of the drug through the BBB.

d) Nanoparticles: Nanoparticles are colloidal systems composed of polymer matrix generally of poly-butyl cyanoacrylate (PBCA). These incorporate drugs within them and are coated with Polysorbate 80 (PS-80). This prevents their sifting by reticuloendothelial system (RES) (Barry et al., 2013). This increases the circulation half-life of drugs.

e) Liposomes: Liposomes are spherical, closed structures which are formed by 1 or more concentric lipid bilayers with an encapsulated aqueous phase in the center and between the bilayers (Bozzuto and Molinari, 2015). Peptides because of their surface charge and large size cannot pass through cell membrane easily. Their repeated injection also leads to immunogenicity. Phospholipids, which are biocompatible, protect the encapsulated peptides from various enzymes present in blood plasma. They also prevent covalent modification (for example by PEGylation) of protein drug thus preventing its loss of activity. They also cause slow release of drugs. Conventional liposomes are sterically stabilized by PEGylation. It increases their hydrophilicity and prevents them from recognition by RES cells such as macrophages. This further helps in increasing their circulation half-life (Bruno et al., 2013)

f) Microspheres: Most commonly prepared microspheres are those of PLGA [Poly (D, L-lactic-co glycolic acid)]. In these microspheres lactic acid and glycolic acid are joined by ester bonds to form the polymer. These ester bonds are hydrolysed by esterases in the blood, once the microspheres are administered to the body (Makadia and Siegel, 2011). Once the drug is released from the polymer, the polymer acids are metabolized and cleared from the body. One of the biggest advantages of microspheres is that they are biocompatible and biodegradable. They provide sustained release of drugs for weeks and months thereby reducing the frequency of administration. One main disadvantage of microspheres is that degradation of PLGA leads to accumulation of lactic and glycolic acids, thus reducing the pH of surrounding environment which causes denaturation of the encapsulated peptide (Pisal et al., 2010).

g) Intranasal administration: One of the best methods to avoid passage through BBB is intranasal administration of drugs. The drug is rapidly absorbed by systemic blood and it does not undergo hepatic metabolism (Misra et al., 2010). Here, the drugs are transported along olfactory sensory neurons and directly enter the CSF. Relative to other strategies this method is non-invasive. This method will have to overcome certain difficulties such as low pH of nasal epithelium which can reduce the activity of drug and the possibility of mucosal irritation (Rahisuddin, 2011).

Conclusion

Neuropeptide research and development is dynamic, with increasing numbers of candidates entering clinical study in a wide variety of therapeutic categories. We anticipate that the pharmaceutical and biotechnology industries will continue to focus on these versatile molecules because of the increased acceptance of injected drugs on the market, the availability of new formulation and delivery technologies, and the relatively high approval success rates. The new and alternative delivery systems open exciting perspectives in the field of neuronal regeneration and neuronal interface, thanks to their unique combination of biological and drug release properties.

References

Aloe L., Rocco M. L., Bianchi P. and Manni L. (2012): Nerve growth factor: from the early discoveries to the potential clinical use. Journal of Translational Medicine, 10: 239.

Barry J. N. and Vertegel A. A. (2013): Nanomaterials for protein mediated therapy and delivery. Nano life, 3(4): 1343001.

Begley D. J. (2003): Understanding and circumventing the blood-brain barrier. Acta Paediatr Suppl. 92(443): 83–91.

Bersani G., Iannitelli A., Fiore M., Angelucci F. and Aloe L. (2000): Data and hypotheses on the role of nerve growth factor and other neurotrophins in psychiatric disorders G. Medical Hypotheses, 55(3): 199–207.

Bozzuto G. and Molinari A. (2015): Liposomes as nanomedical devices. International Journal of Nanomedicine, 10: 975–999.

Brenneman D. E., Phillips T. M., Festoff B. W. and Gozes I. (1997): Identity of neurotrophic molecules released from astroglia by vasoactive intestinal peptide. Annals of New York Academy of Sciences, 814:167-73.

Bruno B. J., Miller G. D. and Lim C. S. (2013): Basics and recent advances in peptide and protein drug delivery. Therapeutic Delivery, 4(11): 1443–1467.

Campos-Salinas J., Cavazzuti A., O' Valle F., Forte-Lago I., Caro M., Beverley S. M. and Gonzalez-Rey E. (2014): Therapeutic Efficacy of Stable Analogues of Vasoactive Intestinal Peptide against Pathogens. The Journal of Biological Chemistry, 289(21): 14583–14599.

Delgado M., Ganea D. (2013): Vasoactive intestinal peptide: a neuropeptide with pleiotropic immune function. Amino acids, 45: 25-39.

Dwibhashyam V. S. N. M. and Nagappa A. N. (2008): Strategies for Enhanced Drug Delivery to the Central Nervous System. Indian Journal of Pharmaceutical Sciences, 70(2): 145–153.

Egleton R. D. and Davis T. P. (2005): Development of neuropeptide drugs that cross blood brain barriers. Neuro Rx, 2: 44-53.

Gonzalez-Rey E., Ganea D. and Delgado M. (2010): Neuropeptides: Keeping the balance between pathogen immunity and immune tolerance. Current Opinion in Pharmacology, 10(4): 473–481.

Gozes I. and Brenneman D. E. (1999): A New Concept in the Pharmacology of Neuroprotection. Journal of Molecular Neuroscience, 14: 61–68.

Gozes I., Divinsky I., Pilzer I., Fridkin M., Brenneman D.E. and Spier A. D. (2003): From vasoactive intestinal peptide (VIP) through activity-dependent neuroprotective protein (ADNP) to NAP: a view of neuroprotection and cell division. Journal of molecular neuroscience, 20(3):315-22.

Hang W. Z. and Liu H. T. (2002): MAPK signal pathways in the regulation of cell proliferation in mammalian cells. Cell Research, 12: 9–18.

Immordino M. L., Dosio F., Cattel L. (2006): Stealth liposomes: review of the basic science, rationale, and clinical applications, existing and potential. Int J Nanomedicine, 1(3): 297-315.

Khan A, Yasir M, Asif M, Chauhan I, Singh A. P., Sharma R, Singh P and Rai S (2011) Iontophoretic drug delivery: History and applications. Journal of Applied Pharmaceutical Science 01 (03): 11-24.

Khan S and Tiwari T (2012): microspheres. World journal of pharmacy and pharmaceutical sciences, 1: 125-145.

Kurakhmaeva K. B., Djindjikhashvili I. A., Petrov V. E., Balabanyan V. U. , Voronina TA, Trofimov S. S., Kreuter J., Gelperina S., Begley D. and Alyautdin R. N. (2009): Brain targeting of nerve growth factor using poly (butyl cyanoacrylate) nanoparticles, Journal of Drug targeting, 8:564-574.

Lodish H, Berk A, Zipursky SL, et al (2000): G Protein –coupled receptors and their effectors in: Molecular Cell Biology, 4th edition, W. H. Freeman, New York.

Makadia H. K., and Siegel S. J. (2011): Poly Lactic-co-Glycolic Acid (PLGA) as Biodegradable Controlled Drug Delivery Carrier. Polymers, 3(3): 1377–1397.

Mikitsh J. L., & Chacko A.-M. (2014): Pathways for Small Molecule Delivery to the Central Nervous System across the Blood-Brain Barrier. Perspectives in Medicinal Chemistry, 6: 11–24.

Misra A., Ganesh S., Shahiwala A. and Shah S. P. (2010): Nasal drug delivery system - an overview. Int. J. Res. Pharm. Sci, 1: 454-465.

Misra A., Ganesh S., Shahiwala A., Shah S. P. (2003): Drug delivery to the Central Nervous System: A review, J Pharma Pharmceuit Sci, 6: 252-173.

Mokhtar S. H., Bakhuraysah M. M., Cram D. S., and Petratos S. (2013): The Beta-Amyloid Protein of Alzheimer's Disease: Communication Breakdown by Modifying the Neuronal Cytoskeleton. International Journal of Alzsheimer's Disease, 2013.

Passemard S., Sokolowska P., Schwendimann L. and Gressens P. (2011): VIP-induced neuroprotection of the developing brain. Current Pharmaceutical Design, 17(10): 1036–1039.

Pathan SA, Iqbal Z, Zaidi SM, Talegaonkar S, Vohra D, Jain GK, Azeem A, Jain N, Lalani JR, Khar RK and Ahmad FJ (2009): CNS drug delivery systems: novel approaches. Recent Pat Drug Deliv Formul., (1):71-89.

Pisal D. S., Kosloski M. P., and Balu-Iyer, S. V. (2010): Delivery of therapeutic proteins. Journal of pharmaceutical sciences, 99(6): 2557–2575.

Prokop, A., and Davidson J. M. (2008): Nanovehicular Intracellular Delivery Systems. Journal of Pharmaceutical Sciences, 97(9), 3518–3590.

Rahisuddin, Sharma P. K., Garg G and Salim M. (2011): Review on nasal drug delivery system with recent advancement. International Journal of Pharmacy and Pharmaceutical Sciences, 3: 611.

Shaji J. and Patole, V. (2008): Protein and Peptide Drug Delivery: Oral Approaches. Indian Journal of Pharmaceutical Sciences, 70(3): 269–277.

Sharma J., Bansal P. K. S., and Banik, A. (2011): Noninvasive Routes of Proteins and Peptides Drug Delivery. Indian Journal of Pharmaceutical Sciences, 73(4): 367–375.

Steingart R. A., Solomon B., Brenneman D. E., and Gozes I. (2000): VIP and Peptides Related to Activity-Dependent Neurotrophic Factor Protect PC12 Cells Against Oxidative Stress. Journal of Molecular Neuroscience, 15: 137–145.1

Upadhyay R. K. (2014). Drug Delivery Systems, CNS Protection, and the Blood Brain Barrier. BioMed Research International, 2014: 869269.

Verge V. M., Richardson P. M., Wiesenfeld-Hallin Z. and Hökfelt T (1995): Differential influence of nerve growth factor on neuropeptide expression in vivo: a novel role in peptide suppression in adult sensory neurons. Neuroscience, 15: 2081-96.

Whitmarsh A. J. (2007): Regulation of gene transcription by mitogen-activated protein kinase signaling pathways. Molecular Cell Research, 1773: 1285-1298.

Wiesmanna I and A.M. de Vos (2000): Nerve growth factor: structure and function. Cellular and Molecular Life Sciences, Mol. Life Sci, 58: 748–759.