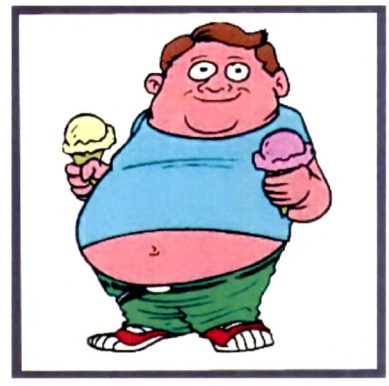
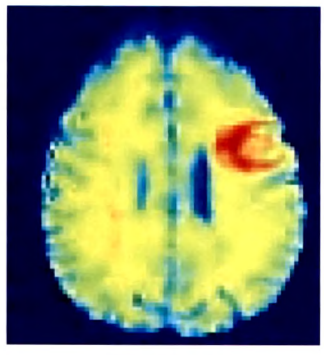


# Chapter 1: Introduction



## 1. INTRODUCTION

The brain is one of the splendorous examples of God's creation controlling all the motor or sensory activities in humans and animals. However, the mechanisms that protect it from exogenous molecules also inhibit the entry of medicaments into the brain, rendering debilitating brain disorders almost untreatable. The treatment of central nervous system (CNS) disorders are challenging because of a variety of formidable obstacles often impede drug delivery to the brain and spinal cord and include the blood brain barrier (BBB) and the blood cerebrospinal fluid barrier (BCF). The blood-brain barriers restrict the passive diffusion of many drugs into the brain and constitute a significant obstacle in the pharmacological treatment of CNS disorders. The efflux mechanisms (P-glycoprotein and MRP, the multi drug resistance-related protein) can also significantly decrease the accumulation of certain drugs within the CNS (Misra et al 2003, Pardridge 2005, Loscher et al 2005). Even though the drugs used for the treatment of CNS disorders are potent, their clinical failure is often not due to lack of drug efficacy but mainly due to short comings in the drug delivery approach. Hence, new strategies for CNS drug delivery and drug targeting are needed.

Many advanced and effective, invasive and noninvasive approaches to the CNS delivery of drugs have emerged in the recent years (Misra et al 2003, Pardridge et al 2005). Intranasal drug delivery is one of the focused delivery option for brain targeting as brain and nose compartments are connected to each other via olfactory/ trigeminal route and peripheral circulation (Illum 2000, Misra et al 2003, Talegaonkar et al 2004, Pardridge et al 2005). Realization of nose to brain transport and the therapeutic viability of the route can be traced from the ancient times and has been successfully investigated for rapid and effective transnasal drug transport in the last two decades. With all its inherent advantages, intranasal route has been indicated as the most promising approach for delivery of drugs to the brain/CNS (Illum 2000, Illum 2003, Talegaonkar et al 2004, Bagger et al 2004). Intranasal administration delivers drug directly to the brain circumventing BBB, and reduces drug delivery to the non-target sites. This results in a reduction in drug dose, systemic dilution and first pass metabolism of the drug (Illum 2000). Direct nose to brain transport results into rapid and higher drug uptake into the brain on intranasal instillation and provides an alternative option of self medication in the management of emergencies. However, the

development of nasal drug products for brain targeting is facing enormous challenges. For overcoming the obstacles, better understanding in terms of factors which are involved in the direct nose to brain transport (physicochemical factors and formulation factors) and transport mechanisms is of utmost importance. Many sophisticated and effective approaches of transnasal drug delivery have emerged in recent years (Arora et al 2002, Vyas et al 2005). Synthesis of more lipophilic analogues, enzyme inhibitors, permeation enhancers, colloidal, bioadhesive and novel drug delivery systems like microemulsion, liposomes and nanoparticles could help in eliminating certain pharmaceutical challenges like low bioavailability, local irritation and toxicity upon long term usage.

The observation that BBB receptor mediated transcytotic systems mediate the brain uptake of circulating endogenous peptides or proteins, such as insulin or transferrin, provided the basis for an approach to brain drug targeting called chimeric peptide technology (Garcia-Garcia et al 2005, Pardridge et al 2005, Tiwari et al 2006). A chimeric peptide is formed when a drug or drug encapsulating nanoconstruct that is not normally transported across the BBB is conjugated to a transport vector also called molecular trojan horse that does undergo transport across the BBB, by means of either RMT for endogenous ligands, or absorptive-mediated transcytosis for cationic proteins or lectins. The transport vector or molecular trojan horse that uses an RMT system at the BBB could be an endogenous peptide, such as insulin or transferrin, or a receptor specific peptidomimetic monoclonal antibody (mAb).

Microemulsions and nanoparticles are widely employed for administering drugs intranasally (LianLi et al 2002, Vila et al 2004, Vyas et al 2005; 2006a; 2006b, Gao et al 2006, Wang et al 2008, Sharma et al 2007, Mistry et al 2009, Sundaram et al 2009, Sharma et al 2009). Nanoparticles and surface modified nanoparticles have been explored as intranasal drug delivery vectors but without any basis and not specifically for nose to brain drug delivery (Gao 2006, Mistry et al 2009, Sundaram et al 2009). Since long, certain receptors have been known to be expressed on the olfactory bulb of nose in comparatively higher abundance than others such as human insulin receptor (HIR) in humans and transferrin receptors in rodents (Hill et al 1985, Hopkins et al 1997, Schulingkamp et al 2000). Therefore, it was hypothesized that a drug loaded nanoconstruct having ligand, specific for these receptors, conjugated to

its surface will be transported selectively to the brain across the olfactory or trigeminal nerves on intranasal delivery through receptor mediated transcytosis (RMT). Also, incorporation of targeting moieties in the nanoparticulate system-improves the therapeutic efficacy, facilitates binding of the conjugates to target cells, promotes conjugate internalization and lowers nonspecific toxicity of the drug.

Moreover, mucoadhesive polymer based drug delivery systems gained importance because of longer residential time of delivery system at the site of administration and possible enhanced transport mechanism by the mucoadhesive agents. Microemulsions have been already explored as an effective carrier system for the rapid and substantial uptake of the drug to the brain (Lianli et al 2002, Vyas et al 2005; 2006a; 2006b). Prolonged residence time of the formulations can be achieved by tailoring the viscosity of the formulation by the addition of various grades and types of mucoadhesive agents. Reports in the literature reveal that chitosan can enhance drug absorption without altering the natural physiological process of the nasal cavity/mucosa. Chitosan have ionic interaction of positive charges of chitosan with negative charges of glycocalix in mucosa.

Despite enormous advances in brain research, brain and central nervous system disorders remain the world's leading cause of disability, and account for more hospitalizations and prolonged care than almost all other diseases combined. Patients suffering from fatal/and/or devastating CNS and associated disorders such as neurodegenerative disorders like Alzheimer's disease, Parkinson's disease, epilepsy, migraine, cerebrovascular diseases and HIV encephalopathy, far outnumber of those victimized from several types of systemic cancers and heart diseases. Beyond the loss of life, this broad category of disorders can have an overwhelming effect on the quality of life for the surviving patient and can lead to serious social and economic burdens on society. Cerebrovascular diseases like cerebral ischemia or stroke can be devastating for patients and their families. Stroke is the second leading cause of death worldwide (Murray et al 1997). Despite the tremendous mortality and morbidity of stroke, treatment options remain limited. However, there is much that can be done to attenuate cerebral damage and reduce the extent of any disability. Active intervention is best seen in three phases: acute therapy, rehabilitation and secondary prevention (Fisher et al 2003). Many pathophysiological key mechanisms of cerebral ischemia

have been identified in recent years, but drug treatment targeting one or a few of these mechanisms has failed to improve clinical outcome after stroke. The most plausible reason for this failure is the multiplicity of mechanisms involved in causing neuronal damage during ischemia. Drugs targeting a multimodal mode of action could potentially overcome this dilemma, and have recently been shown to provide a remarkable benefit in preclinical studies. Till date there is no effective approved therapy to manage cerebral ischemia or stroke and its consequences. At present the only FDA approved treatment is to provide tissue plasminogen activator to reopen occluded blood vessels, however, due to a narrow time window, this treatment is only appropriate for a very small number of patients. Antiplatelet, antihypertensive, and lipid-lowering therapies are approved for secondary stroke prevention. The therapeutics selected for the study i.e hydergine and nicergoline may prove to be an effective treatment for cerebral ischemia following intranasal administration, as will exert their anti-ischemic activity by virtue of vasodilatation, anti-platelet aggregability and antioxidant properties (Nishio et al 1998, Sozmen et al 1998, Vairetti et al 2002, Vairetti et al 2004).

Obesity is a growing health problem in many of the richest nations of the world and is now considered a chronic disease that is reaching epidemic proportions (Barness et al 2007). Obesity in general is defined as a state of 'excess body fat' or 'body weight that is 20% over the ideal' and is expressed in terms of 'Body Mass Index' (BMI). Obesity increases the likelihood of various diseases, particularly heart disease, type 2 diabetes, breathing difficulties during sleep, certain types of cancer, and osteoarthritis (Haslam et al 2005). It has now been explored that the body has a homeostatic mechanism for controlling body fat and that the central nervous system (CNS) is involved. Many reviews of energy balance control contain spaghetti diagrams of interacting factors endocrines, autonomic mediators, gastrointestinal peptides, CNS transmitters, etc all impinging on the hypothalamus, which, in turn, releases mediators that act on CNS, autonomic and endocrine systems affecting food intake and energy balance (Mantzoros 1999, Flier 2004). The anti-obesity drug selected for the study, sibutramine, was a clinically approved orally administered, centrally acting anti-obesity drug and produced its therapeutic effects by norepinephrine, serotonin and dopamine reuptake inhibition. It has been withdrawn due to serious peripheral side

effects such as cardiovascular disturbances, gastrointestinal disturbances, etc arising from unwanted tissue distribution (Heal et al 1998).

In the light of above facts, an alternative drug delivery system is needed which can selectively target the candidate drugs to the various regions of brain. Due to improved transport of drugs to the brain, intranasal delivery approach may be expected to reduce the wide distribution of drug to the non-targeted sites such as systemic/ peripheral circulation. The delivery system must be meticulously designed to provide preferential and rapid transport of drug across nasal mucosa.

## RESEARCH ENVISAGED

Hence the aim of this investigation was envisaged to deliver nicergoline, hydergine and sibutramine via nasal route for the effective treatment of CNS disorders like cerebral ischemia and obesity. It was hypothesized that an intranasal monoclonal antibody conjugated drug containing nanoparticulate formulation will selectively and effectively deliver drugs to the brain, and will result in reduction of the dose of the drug and drug associated serious systemic side effects by delivering drug directly to the target organ and minimizing systemic exposure of the drug.

The proposed plan of research includes:

1. Review of literature with reference to CNS disorders, cerebral ischemia or stroke and obesity, intranasal delivery for brain targeting, delivery system based approaches for intranasal delivery of drugs, nanoparticles, microemulsion, mucoadhesive agents, nasal gel, radiolabeling, analytical profile and physicochemical properties of the selected therapeutic agents.
2. Preparation of solutions containing selected drugs, preparation, optimization and characterization of unconjugated and antibody conjugated nanoparticles with the help of factorial designing and evaluation of stability of the formulations.
3. Preparation of solutions containing selected drugs, preparation, optimization and characterization of microemulsions and mucoadhesive microeulsions with the help of factorial designing and evaluation of stability of the formulations.

4. Radiolabeling of the selected formulations and optimization of radiolabeled complex for its suitability for *in vivo* studies.
5. Biodistribution studies and gamma scintigraphy imaging in animals to ascertain nose to brain transport of drug.
6. Pharmacodynamic studies of the drugs on suitable animal models (high fat diet induced obese animals for obesity and transient global ischemic model for cerebral ischemia).

## REFERENCES


- Arora P, Sharma S, Garg S. Permeability issues in nasal drug delivery. *Drug Discov Today*, 2002, 7(18), 967-975.
- Bagger MA, Bechgaard E. The potential of nasal application for delivery to the central brain-a microdialysis study of fluorescein in rats. *Eur J Pharm Sci*, 2004, 21(2-3), 235-42.
- Barness LA, Opitz JM, Gilbert-Barness E. Obesity: genetic, molecular, and environmental aspects. *Am J Med Genet A*, 2007, 143A, 3016-3034.
- Fisher M, Brott TG. Emerging Therapies for Acute Ischemic Stroke-New Therapies on Trial. *Stroke*, 2003, 34, 359-361.
- Flier JS. Obesity wars: Molecular progress confronts an expanding epidemic. *Cell*, 2004, 116, 337-350.
- Gao X, Tao W, Lu W, *et al.* Lectin-conjugated PEG-PLA nanoparticles: preparation and brain delivery after intranasal administration. *Biomaterials,* 2006, 27, 3482-3490.
- Garcia-Garcia E, Andrieux K, Gilb S, Couvreur P. A Review Colloidal carriers and blood–brain barrier (BBB) translocation: A way to deliver drugs to the brain? *Int J Pharm*, 2005, 298, 274-292.
- Haslam DW, James WP. Obesity. *Lancet*, 2005, 366, 1197-1209.
- Heal DJ, Aspley S, Prow MR, Jackson HC, Martin KF, Cheetham SC. Sibutramine: a novel anti-obesity drug. A review of the pharmacological evidence to differentiate it from d-amphetamine and d-fenfluramine. *Int J Obes Relat Metab Disord,* 1998, 22 Suppl 1, S18-28, discussion S29.
- Hill JM, Ruff MR, Weber RJ, Pert CB. Transferrin receptors in rat brain: neuropeptide-like pattern and relationship to iron distribution. *Proc Natl Acad Sci USA*, 1985, 82, 4553-4557.

- Hopkins DFC, Williams G. Insulin receptors are widely distributed in human brain and bind human and porcine insulin with equal affinity. *Diabet Med*, 1997, 14, 1044-1050.
- Illum L. Transport of drugs from the nasal cavity to the central nervous system. *Eur J Pharm Sci*, 2000, 11,1-18.
- Illum, L. Nasal drug delivery-possibilities, problems and solutions. *J Control Release*, 2003, 87(1-3), 187-198.
- Lian Li, Nandi I, Kim KH. Development of an ethyl laurate-based microemulsion for rapid-onset intranasal delivery of diazepam. *Int J Pharmaceutics,* 2002, 237(1-2), 77-85.
- Loscher W, Potschka H. Role of drug efflux transporters in the brain for drug disposition and treatment of brain diseases. *Prog. Neurobiol.*, 2005, 76(1), 22-76.
- Mantzoros CS. The role of leptin in human obesity and disease: a review of current evidence. *Ann Intern Med*, 1999, 130, 671-680.
- Misra A, Ganesh S, Shahiwala A, Shah SP. Drug delivery to the central nervous system: a review. *J Pharm Pharmaceut Sci,* 2003, 6(2), 252-273.
- Mistry A, Stolnik S, Illum L. Nanoparticles for direct nose-to-brain delivery of drugs. *Int J Pharm,* 2009, 379, 146-157.
- Murray CJ, Lopez AD. Mortality by cause for eight regions of the world: Global Burden of Disease Study. *Lancet*, 1997, 339, 342-344.
- Nishio T, Sunohara N, Furukawa S, Akiguchi I, Kudo Y. Repeated injections of nicergoline increase the nerve growth factor level in the aged rat brain. *Jpn J Pharmacol.*, 1998, 76(3), 321-323.
- Pardridge WM. The blood-brain barrier: Bottleneck in brain drug development. *NeuroRx*, 2005, 2, 3-14.
- Schulingkamp RJ, Pagano TC, Hung D, Raffa RB. Insulin receptors and insulin action in the brain: review and clinical implications. *Neurosci Biobehav Rev,* 2000, 24, 855-872.
- Sharma BV, Ali M, Baboota S, Ali J. Preparation and characterization of chitosan nanoparticles for nose to brain delivery of a cholinesterase inhibitor. *Indian J Pharm Sci*, 2007, 69, 712-713.
- Sharma G, Mishra AK, Mishra P, Misra A. Intranasal cabergoline: pharmacokinetic and pharmacodynamic studies. *AAPS PharmSciTech*, 2009, 10, 1321-1330.

- Sozmen EY, Kanit L, Kutay FZ, Hariri NI. Possible supportive effects of co-dergocrine mesylate on antioxidant enzyme systems in aged rat brain. *Eur Neuropsychopharmacol.*, 1998, 8(1), 13-16.
- Sundaram S, Roy SK, Ambati BK, Kompella UB. Surface-functionalized nanoparticles for targeted gene delivery across nasal respiratory epithelium. *FASEB J,* 2009, 23, 3752-3765.
- Talegaonkar S, Mishra PR. Intranasal delivery: An approach to bypass the blood brain barrier. *Indian J Pharmacol,* 2004, 36(3), 140-147.
- Tiwari SB, Amiji MM. A review of nanocarrier-based CNS delivery systems. *Curr Drug Deliv*, 2006, 3, 219-232.
- Vila A, Sanchez A, Janes K, *et al.* Low molecular weight chitosan nanoparticles as new carriers for nasal vaccine delivery in mice. *Eur J Pharm Biopharm,* 2004, 57, 123-131.
- Vyas TK, Shahiwala A, Marathe S, Misra A. Intranasal Drug Delivery for Brain Targeting. *Curr Drug Deliv,* 2005, 2(2), 165-175.
- Vyas TK, Babbar AK, Sharma RK, Misra A. Intranasal mucoadhesive microemulsions of zolmitriptan: preliminary studies on brain-targeting. *J Drug Target,* 2005, 13, 317-324.
- Vyas TK, Babbar AK, Sharma RK, Shashi S, Misra A. Intranasal mucoadhesive microemulsions of clonazepam: preliminary studies on brain targeting. *J Pharm Sci,* 2006, 95, 570-580.
- Vyas TK, Babbar AK, Sharma RK, Misra A. Preliminary brain targeting studies on intranasal mucoadhesive microemulsions of sumatriptan. *AAPS PharmSciTech,* 2006, 7, E8.
- Wang X, Chi N, Tang X. Preparation of estradiol chitosan nanoparticles for improving nasal absorption and brain targeting. *Eur J Pharm Biopharm,* 2008, 70, 735-740.
- Vairetti M, Battaglia A, Carfagna N, Luigi Canonico P, Berte F, Richelmi P. Antioxidant properties of MDL and MMDL, two nicergoline metabolites, during chronic administration of haloperidol. *Eur J Pharmacol.*, 2002, 453(1), 69-73.
- Vairetti M, Ferrigno A, Canonico PL, Battaglia A, Berte F, Richelmi P. Nicergoline reverts haloperidol-induced loss of detoxifying-enzyme activity. *Eur J Pharmacol.*, 2004, 505(1-3), 121-125.