Chapter 2
Literature Review

2.1 NASAL DRUG DELIVERY

The interest in and importance of the systemic effects of drugs administered via the nasal route have expanded over recent decades. The schematic of nasal drug delivery is shown in Fig. 2.1. Nasal administration offers an interesting alternative for achieving systemic drug effects to the parenteral route, which can be inconvenient or oral administration, which can result in unacceptably low bioavailabilities (Harris, 1993). The nasal epithelium is a highly permeable monolayer, the submucosa is richly vascularised, and hepatic first-pass metabolism is avoided after nasal administration. Other attractive features include the rather large surface area of the nasal cavity and the relatively high blood flow, which promotes rapid absorption (Chien et al., 1989). Furthermore, self-medication is easy and convenient.

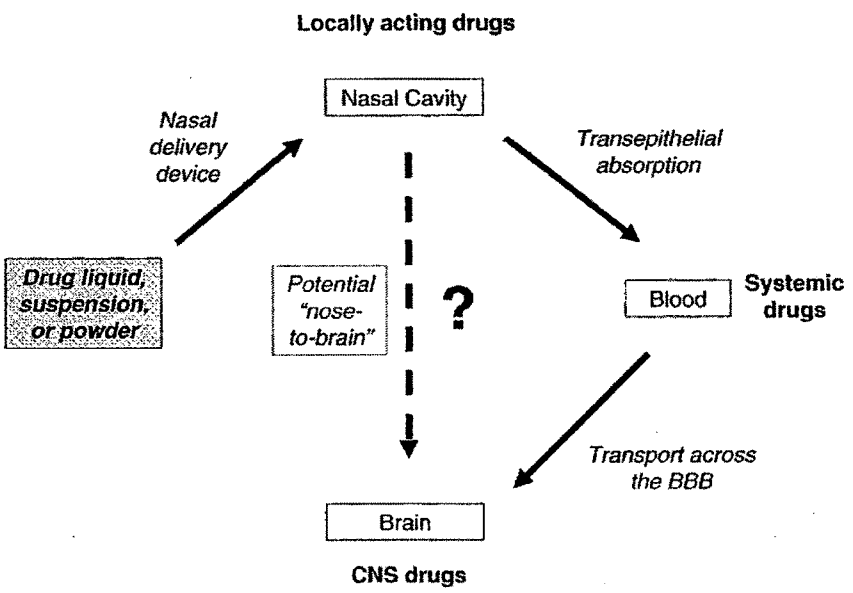


Fig. 2.1 Schematic of nasal drug delivery. IN drugs formulated as solutions, suspensions, or powders can be administered to the nasal cavity (local action), can transport across the epithelial tissue to enter the blood (systemic drugs), and for drugs that cross the blood–brain barrier (BBB), can subsequently enter the brain (CNS applications). Direct delivery of IN drugs to the brain has been proposed, but is not universally established in the literature (Costantino et al., 2007).

The nasal cavity offers a number of unique advantages such as easy accessibility, good permeability especially for lipophilic, low molecular weight drugs, avoidance of harsh environmental conditions and hepatic first pass metabolism, potential direct delivery to the brain, and direct contact for vaccines with lymphatic tissue and action as inducer as well as effector of the mucosal immune system. The nasal epithelium is well suited for the transmucosal drug delivery although it is less permeable for hydrophilic and high molecular weight drugs. Ciliary movement and the resulting clearance of the delivered drug / dosage form towards the throat are challenges when developing a prolonged release dosage form. Also a considerable enzyme activity, though lower than in the gastrointestinal tract, must be considered. Nevertheless, a number of approaches have been used to overcome these limitations such as the use of mucoadhesive formulations to increase the nasal residence time of dosage forms (Morimoto et al., 1991; Soane et al., 2001), addition of absorption enhancers to increase the membrane permeability (De Ponti, 1991; Merkus et al., 1993; Illum, 1999; Natsume et al., 1999), and the use of protease / peptidase inhibitors to avoid enzymatic degradation of peptide and protein drugs in the nasal cavity (Morimoto et al., 1995; Dondeti et al., 1996). Several nasal dosage forms are under investigation including solutions (drops or sprays), gels, suspensions and emulsions, liposomal preparations, powders and microspheres, as well as inserts.

2.1.1 The nasal cavity

The nose as a drug delivery site has a number of unique features related to its anatomy and physiology. These features have to be taken into consideration when developing a nasal drug delivery system. The following sections will therefore give an introduction to the anatomy and physiology of the human nose.

2.1.2 Nasal Anatomy

The nose is part of the upper respiratory system and is the main route by which ambient air enters the body. The apparent external nose surrounds the nostrils and one third of the nasal cavity. The entire human nasal cavity is an approximately 5 cm high and 10 cm long dual chamber with a total surface area of about 150 cm^2 and a total volume of about 15 - 20 ml. The nasal cavity is divided by the nasal septum into two halves of approximately equal size, beginning anteriorly at the nares and extending posteriorly to the nasopharynx where the two halves of the airway join together.

Located approximately 1.5 cm from the nares is the narrowest portion of the entire airway, the internal ostium (or nasal valve) with a cross-sectional area of about 30 mm^2 on each side (Fig. 2.2). The nasal valve accounts for approximately 50% of the total resistance to respiratory airflow from the nostrils to the alveoli (Mygind and Dahl, 1998). This high resistance to airflow, the relatively high linear velocity of the air stream, combined with an almost 90° angle of the flow passage at the ostium, and turbulences facilitate the impaction of the majority of particles carried in the inspired air stream in the anterior of the nasal cavity from where they are mainly removed by mucociliary clearance (Hinchcliffe and Illum, 1999).

Each half of the nasal cavity is limited by the septal wall and the lateral wall. Bony scroll-like conchae (or turbinates) are attached to the lateral wall and project into the main part of the cavities (Fig. 2.2). Although more complex in many animal species, in human three conchae, called the inferior, median, and superior, have a relatively simple scroll arrangement, (Gizurarson, 1990; Illum, 1996). The presence of these chonchae creates a turbulent air flow through the nasal passages which ensures a better contact between the mucosa and the inspired air, thus facilitating its humidification and temperature regulation.

Underneath and lateral to each of the turbinates are passages called the inferior, middle, and superior meatus. The inferior and middle meatus receive the openings of the nasolachrymal duct and the paranasal sinuses. The mucous membrane in a meatus will not be hit by an ordinary nasal spray (Mygind and Dahl, 1998).

In the posterior part of the nasal cavity the air passage bends again about 90° as it passes to the nasopharynx and the air stream increases in velocity, resulting in impaction of particles in the posterior of the nasal cavity from where they are removed by mucociliary clearance.

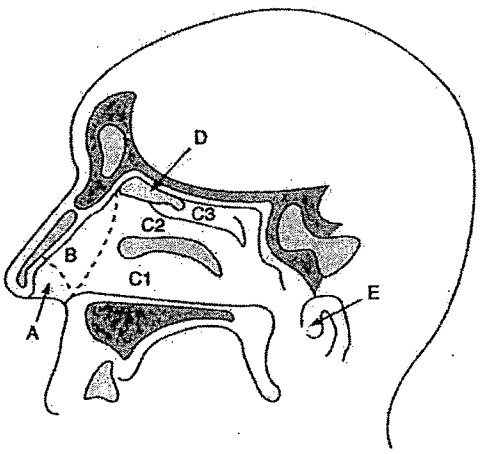


Fig. 2.2 Schematic of a sagittal section of human nasal cavity showing the nasal vestibule (A), atrium (B), respiratory region: inferior turbinate (C1), middle turbinate (C2) and the superior turbinate (C3), the olfactory region (D) and nasopharynx (E) (Ugwoke et al., 2001).

2.1.3 Nasal Physiology

According to their function, the nasal cavity can be divided into three regions (Merkus and Verhoef, 1994):

(i) The **vestibular region** (the anterior 10 - 20 cm^2) is posteriorly limited by the internal ostium. It is covered with a stratified squamous epithelium, which is continuous with the facial skin. Short stiff hairs filter larger particles from the incoming air stream.

(ii) The **respiratory region** (about 130 cm^2) occupies the majority of the main part of the nasal cavities (posterior two thirds) and is important for the absorption of drugs into the systemic circulation. The epithelium consists of pseudostratified columnar epithelial cells.

(iii) The **olfactory region** (10 - 20 cm^2) at the roof of the nasal cavities comprises of the small patch of columnar cells containing the smell receptors (Fig. 2.2).

2.1.3.1 Respiratory surface epithelium:

The respiratory epithelium has a thickness of approximately 100 µm (Merkus and Verhoef, 1994). It consists of following major cell types (Fig. 2.3) (Ugwoke et al., 2001).

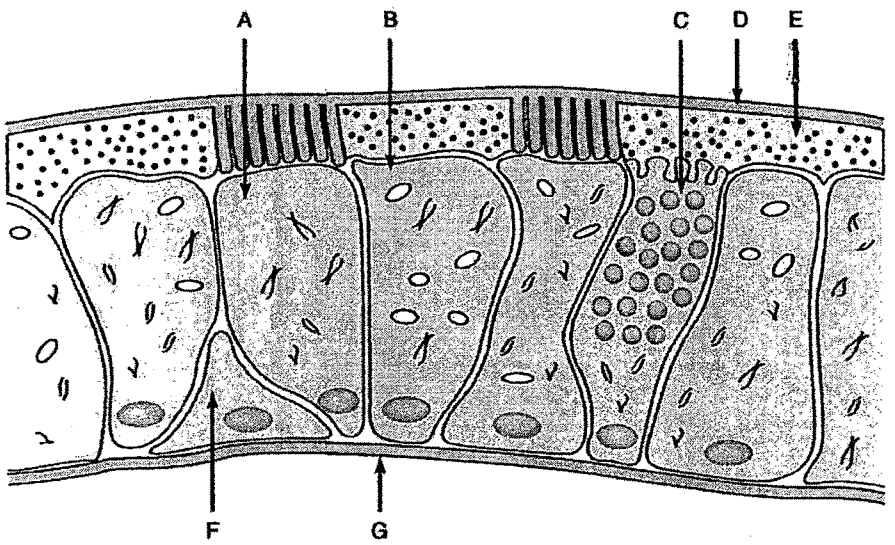


Fig. 2.3 Cell types of the nasal epithelium showing ciliated cell (A), non-ciliated cell (B), goblet cells (C), gel mucus layer (D), sol layer (E), basal cell (F) and basement membrane (G) (Ugwoke et al., 2001).

(i) Basal cells, which are progenitors of the other cell types, lie on the basement membrane and do not reach the airway lumen.

(ii) Columnar cells are related to neighboring cells by tight junctions. The cytoplasm contains numerous mitochondria in the apical part, as a sign of an active metabolism. All columnar cells are covered by about 300 microvilli, uniformly distributed to the entire apical surface. These short and slender fingerlike cytoplasmic expansions increase the surface area of the epithelial cells, thus promoting exchange processes across the epithelium. The microvilli also prevent drying of the surface by retaining moisture essential for ciliary function. Columnar cells can be divided into non-ciliated and ciliated cells. Cilia are fingerlike protrusions (0.2 - 0.3 μm wide and 5 μm in length) on the apical surface of cells, which have a typical ultrastructure and are larger than microvilli. Each ciliated cell contains about 100 - 300 cilia (Petruson et al., 1984). The anterior third of the nasal cavity is non-ciliated. Cilia start occurring just behind the front edge of the inferior turbinate. The posterior part of the nasal cavity as well as the paranasal sinuses are densely covered with cilia.

(iii) Goblet cells are mucous-containing and secreting cells typical for a respiratory epithelium. Their number is slightly larger in the posterior than in the anterior part of the nasal cavity with a mean concentration of goblet cells (4,000 - 7,000 / mm^2)

similar to the trachea and the main bronchi (Tos, 1983). The contribution of goblet cells to the volume of nasal secretion is probably small compared to that of the submucosal glands.

Little is known about the release mechanism from goblet cells, which in contrast to submucosal glands are not under parasympathetic control. Goblet cells probably respond to physical and chemical irritants in the microenvironments.

2.1.3.2 Submucosal glands, mucus, and mucociliary clearance:

Below the respiratory epithelium is a thick lamina propria, composed of a loose mesh of fibroelastic tissue with many blood vessels, nerves, and glands. These submucosal glands possess both serous and mucous secretory cells and release directly onto the surface of the epithelium. The majority of what is referred to as 'nasal secretion' is produced by the glands. Other minor contributors are goblet cells and plasma exudation, especially during inflammatory processes.

A thin, clear, and continuous layer of fluid, called mucus, covers the entire nasal epithelial surface. Approximately 20 - 40 ml of mucus is produced from the normal 'resting' nose each day (Quraishi, 1998). This mucus is composed of water (95 - 97%), mucus glycoproteins (2.5 - 3%), electrolytes (1%), proteins (1%), and other macromolecules (Kaliner et al., 1984). The baseline pH in the human nasal cavity is approximately 6.3, ranging from 5.2 - 8.1 (Washington et al., 2000). The mucus glycoproteins (mucins) consist of a protein core (20%) with oligosaccharide side chains (80%), cross-linked by disulfide and hydrogen bonds (Kaliner et al., 1984). These glycoproteins are responsible for the characteristic viscoelastic properties of the mucus, which are related to its function of providing a protective coating to the nasal epithelium and mucociliary clearance. Mucus consists of two fluid layers, each approximately 5 μm thick: a viscous gel layer (mucus or epiphase) floats on a less viscous sol layer (periciliary fluid or hypophase) immediately adjacent to the epithelial surface. The cilia of the columnar cells move with regular, symmetric beats at a frequency of about 10 Hz in the lower sol phase (Duchateau et al., 1985). During this process, the ciliary tips make contact with and propel the gel layer whilst the sol layer remains relatively stationary (Sleigh et al., 1988). During the recovery stroke the cilia move backward exclusively through the sol layer.

By this action the upper mucus layer, together with deposited particles, is transported towards the nasopharynx from where it is swallowed. The velocity of mucous transport is approximately 5 - 8 mm/min (Procter et al., 1973; Andersen and Procter, 1983), thus renewing the nasal mucus layer every 10 - 20 min. The combined action of mucus layer and cilia is called mucociliary clearance. It is an important nonspecific physiological defense mechanism of the respiratory tract to protect the body against noxious inhaled materials. Inhibition of the mucociliary clearance by drugs and drug delivery systems results in longer contact times of the nasal mucosa with inhaled bacteria, viruses, carcinogens etc. On the other hand, the mucociliary clearance is responsible for the generally observed rapid clearance of nasally administered drugs from the nasal cavity to the nasopharynx. It forms, therefore, an opposing mechanism in the absorption process of drugs following intranasal delivery. To overcome the rapid removal of nasally administered drugs the concept of mucoadhesion can be applied.

2.1.3.3 Vasculature and innervation:

The lamina propria under the nasal epithelium and the basement membrane is rich in blood vessels and has an extensive blood supply (about 40 ml/min/100g) (Bende et al., 1983) as well as a large lymph drainage system, particularly in the respiratory region of the nasal cavity (Hinchcliffe and Illum, 1999). These blood vessels differ from the vasculature of the tracheobronchial tree in three ways (Mygind and Dahl, 1998):

(i) Cavernous venous sinusoids are specialized vessels adapted to the functional demands of the nose with respect to heating and humidification of inhaled air. When they distend with blood the mucosa will swell and block the airway lumen.

(ii) Arterio-venous anastomoses allow the blood to bypass the capillaries. Their role is probably related to the temperature and water control. At least 50% of the blood flow is normally shunted through arterio-venous anastomoses (Anggard, 1974).

(iii) Nasal vasculature shows cyclic changes of congestion (nasal cycle, every 3 – 7 h).

Engorgement of venous plexuses with blood leads to a swelling of the mucosa which can temporarily occlude the airway and make the tissue appear erectile. This

occlusion of the airway is thought to occur alternately between the two sides of the nasal cavity preventing the drying-out of the mucous membrane (Hinchcliffe and Illum, 1999). The effect of the nasal cycle on the absorption of nasally administered drugs is still unclear. The total clearance of radiolabelled saline was not affected by the nasal patency, even though the initial clearance was higher in the more patent half of the nasal cavity (Washington et al., 2000).

Different to the gastrointestinal tract, the venous blood draining from the nose passes directly into the systemic circulation, thereby circumventing hepatic first pass elimination. The lamina propria of the nasal mucosa embeds also nerves. Afferent nerve fibers run in the trigeminal nerve. Stimulation of the trigeminus in the nasal mucosa results in the sneezing reflex. There is a rich parasympathetic innervation of the glands. Nervous stimulation of the glandular cholinoceptors causes marked hypersecretion and is often part of the reflex arc. Nasal blood vessels are both sympathetically and parasympathetically innervated, but are mainly controlled by sympathetic fibers (Mygind and Dahl, 1998).

2.2 Function of the nose:

Humans breathe in about 12 - 24 times/min, thereby inhaling daily approximately 10,000 liters of air of differing temperature and humidity, containing dust and organisms. As the main entrance for inspired air, the nose has the following functions (Jones, 2001):

(i) Olfaction
(ii) Sensation
(iii) Immunology
(iv) Clearing of inspired air
(v) Heating and humidification of inspired air
(vi) Resonance organ and isolation

It is without question that administration of drugs via the nasal route and nasally applied drug delivery systems should not interfere with these physiological functions of the nose.

2.3 Nasal metabolism:

Absorption of drugs across the nasal mucosa results in direct systemic exposure, thus avoiding hepatic first-pass metabolism associated with oral administration. However, an alternative first-pass effect is created by the metabolic activity in the nasal mucosa. Metabolizing enzymes are present in nasal secretions, in nasal epithelial cells (in the cytosol and membrane-bound) and in the lamina propria (Hinchcliffe and Illum, 1999). Although much of the literature concerning nasal enzymes relates to studies in animals (rat, rabbit, Syrian hamster, dog, and monkey), the profile of nasal enzymes in humans is considered similar, despite inter-species variations (Sakar, 1992).

Monooxygenases, reductases, transferases, esterases and proteolytic enzymes were identified in the nasal mucosa (Irwin et al., 1995). Oxidative phase I enzymes such as cytochrome P-450 dependent monooxygenase are recognized as a potential first line defense of the upper respiratory tract against airborne xenobiotics. Numerous compounds are metabolized in vitro by nasal cytochrome P-450, e.g. nasal decongestants, essences, anesthetics, alcohols, nicotine, and cocaine (Sakar, 1992). The specific content of P-450 in the nasal mucosa is relatively high, second only to that of the liver. The catalytic activity is more pronounced in the olfactory region of the nose than in the respiratory region (Brittebo, 1982). Also phase II enzymes such as glutathione transferase are present in the nasal mucosa.

Alternative mucosal routes of administration, such as the nasal mucosa, are especially interesting with regards to protein and peptide delivery. However, the nasal epithelium and the nasal secretions contain various peptidase and protease activities, including exopeptidases as well as endopeptidases (Lee and Yamamoto, 1990). Aminopeptidases are the principle proteolytic enzymes in the nasal mucosa (Kashi and Lee, 1986; Audus and Tavakoli-Saberi, 1991), with almost half of the aminopeptidase activity being membrane bound (Lee and Yamamoto, 1990). Inhibition of proteolytic enzymes is also discussed as a contributing mechanism for some penetration enhancers, e.g. sodium glycocholate, which has an inhibitory effect on aminopeptidase activity (Hirai et al., 1981).

In general, nasal administration of drugs has to consider a pseudo-first-pass effect caused by enzymes in the nasal cavity. Naturally the metabolic clearance of substances from the nose into the blood is highly variable, depending on the particular

compound under investigation. Whether a nasally administered drug is subject to a nasal first-pass metabolism depends on the presence of specific isozymes and the contact time. Propranolol for instance, a drug which suffers extensive gastrointestinal and hepatic first-pass metabolism after oral administration (Walle et al., 1985) is rapidly absorbed unmetabolized after nasal administration (Hussain et al., 1980).

2.4 The nose as drug delivery site: advantages, barriers, and solutions

The nose as a site of drug administration offers the following **advantages**:

(i) Easy accessibility and needle free drug application without the necessity of trained personnel facilitates self-medication, thus improving patient compliance compared to parenteral routes (Pontiroli et al., 1989).

(ii) Good penetration of, especially lipophilic, low molecular weight drugs through the nasal mucosa.

(iii) Rapid absorption and fast onset of action due to a relatively large absorptive surface and high vascularization. Nasal administration of suitable drugs would therefore be effective in emergency therapy as alternative to parenteral administration routes.

(iv) Avoidance of the harsh environmental conditions in the gastrointestinal tract (chemical and enzymatic degradation of drugs).

(v) Avoidance of hepatic first-pass metabolism and thus potential for dose reduction compared to oral delivery.

(vi) Potential for direct delivery of drugs to the central nervous system via the olfactory region under bypassing the blood-brain-barrier (Illum, 2000; Chow et al., 2001; Dahlin et al., 2001; Dufes et al., 2003).

(vii) Direct delivery of vaccine to lymphatic tissue and secretory immune response at distant mucosal sites.

Despite these advantages of the nose as drug delivery site, certain **barriers** may be encountered when developing a nasal drug formulation:

(i) Bioavailabilities of polar drugs are generally low, about 10% for low molecular weight drugs and not above 1% for peptides such as calcitonin and insulin (Illum, 2003). The most important factor limiting the nasal absorption of polar drugs and especially large molecular weight polar drugs such as peptides and proteins is the low membrane permeability. Drugs can cross the epithelial cell membrane either by the transcellular route exploiting simple concentration gradients, by receptor mediated or vesicular transport mechanisms, or by the paracellular route through the tight junctions between the cells. Polar drugs with molecular weights below 1000 Da will generally pass the membrane using the latter route (McMartin et al., 1987). Although tight junctions are dynamic structures and can open and close to a certain degree when needed, the mean size of the channels is of the order of less than 10 Å and the transport of larger molecules is considerably more limited (Madara and Dharmsathaphorn, 1985; McMartin et al., 1987).

(ii) The general fast clearance of the administered formulation from the nasal cavity due to the mucociliary clearance mechanism is another factor of importance for low membrane transport. This is especially the case when the drug is not absorbed rapidly enough across the nasal mucosa. It has been shown that for both liquid and powder formulations, which are not bioadhesive, the half life for clearance is of the order of 15 - 30 min (Illum et al., 1987; Soane et al., 1999, Soane et al., 2001). The use of mucoadhesive excipients in the formulations is an approach to overcome the rapid mucociliary clearance. The clearance may also be reduced by depositing the formulation in the anterior, less ciliated part of the nasal cavity thus leading to improved absorption (Harris et al., 1986; Kublik and Vidgren, 1998).

(iii) Another contributing, but often less considered factor to the low bioavailability of peptides and proteins across the nasal mucosa is the possibility of an enzymatic degradation of the molecule in the lumen of the nasal cavity or during passage through the epithelial barrier. The use of enzyme inhibitors and / or saturation of enzymes may be approaches to overcome this barrier (Morimoto et al., 1995).

In summary, the nose offers unique advantages as administration site for drug delivery. However, low permeability for polar and high molecular weight drugs, rapid clearance of the delivery system from the cavity and possible enzymatic degradation

of the drug in the nose may be encountered. These challenges can be faced by various approaches, such as use of bioadhesive systems and absorption enhancers.

Due to the pronounced advantages, both small molecules and macromolecules are extensively studied for nasal delivery, as listed in Table 2.1.

Table 2.1 Small molecules and macromolecules currently being studied for nasal delivery (Talegaonkar and Mishra, 2004)

	Small molecules	*Macro molecules*
Adreno corticosteroids		
Antibiotics	Gentamicin, Cephalosporin, Penicillins, Tyrothricin	Amino acids-
Antimigraine drugs	Dihydroergotamine, Ergotamine tartrate	**Peptides**
Antiviral drug	Enviroxime	Calcitonin, Secretin,Thyrotropin-releasing
Cardiovascular drugs	Isosorbide dinitrate, Propranolol, Verapamil, Hydralazine, Nitroglycerin, Clofillium tosylate	hormone (TRH), Cerulein, Enkephalin analogs- Leucine enkephalin, Mekephamid
Central nervous system drugs		Pentagastrin, SS-6, Substance P, Kyotorphin,
a. Stimulants	Cocaine, Lidocaine	Cholecystokinin
b. Depressants	Diazepam, Lorazepam	
Autonomic nervous system drugs		
a. Sympathomimetics	Dopamine, Dobutamine, Ephedrine, Epinephrine, Phenyle phrine, Tramazoline, Xylometazoline	**Polypeptides and proteins**
b. Parasympathomimetics	Methacholine, Nicotine	a. Albumins
c. Parasympatholytics	Atropine, Prostaglandins, Ipratropium, Scopolamine	b. Anterior pituitary hormones - Adreno-
Diagnostic drugs	Dye T-1824, Phenolsulfonphthalein, Potassium ferrocyanide, Vital dyes	corticotropic hormone, Gonadotropin-releasing hormone, Growth hormone
Histamine and antihistamines		c. Biological products - Interferon, Vaccines
a. Histamine		d. Horseradish peroxidase
b. Antihistamines-	Medizine	e. Pancreatic hormones - Insulin, Glucagon
c. Mast cell stabilizers -	Disodium cromoglycate	f. Posterior pituitary hormones - Oxytocin,
Narcotics and antagonists	Buprenorphine, Naloxone	Vasopressin
Sex Hormones	Estradiol, Progesterone, Norethindrone, Testosterone	
Inorganic compounds	Colloidal carbon, Colloidal gold, Inorganic salts, Lead carbonate P and Thorium B	
Vitamins	Folic acid, Cyanocobalamin	

2.5 Intranasal Drug Absorption

To reach the vascular system, substances intended for systemic absorption must first pass through the mucus layer followed by the epithelium. A review by Khanvilkar et al. (Khanvilkar et al. 2001) addresses the numerous issues associated with drug transfer through mucus. The mucus does not present a major problem for small, uncharged particles. On the other hand, some larger or charged molecules may encounter difficulties passing through this layer. One important rate-limiting factor in the diffusion of drug through the mucus is potential binding of solutes to mucin. Types of interactions between foreign molecules and the mucus include electrostatic, hydrophobic, and van der Waals forces. In addition, the mucus' structure is very sensitive to its environment, meaning that alterations in pH, temperature, osmotic pressure, etc. may induce structural changes in this layer. This dynamic nature of the

mucus may cause variations in the transfer of molecules from the delivery site to the epithelium (Khanvilkar et al. 2001).

Once a substance passes through the mucus, it may cross the nasal mucosa by three different mechanisms (Fig. 2.4) (Martin et al., 1997). First, it may be transferred via transcellular, or simple, diffusion across the membrane by way of pores or carriers (Lee, 1988). A second method is paracellular transport, which involves movement through the spaces between cells and tight junctions (Lee, 1988). This pathway is believed to be important for absorption of peptides and proteins (Agarwal and Mishra, 1987). The third mechanism is transcytosis. This entails particle uptake into vesicles, subsequent transfer across the cell, and, finally, deposition into the interstitial space (McMartin et al., 1987).

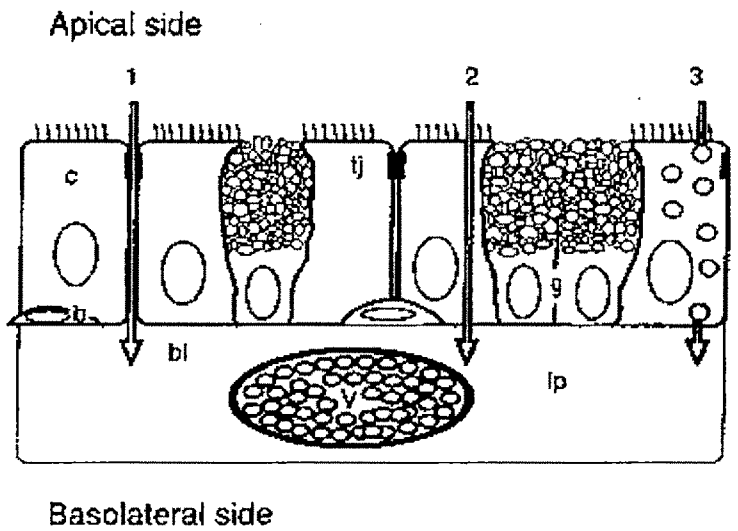


Fig. 2.4 Schematic representation of transport routes across nasal respiratory epithelium: (1) paracellular; (2) transcellular; (3) transcytotic. Goblet cells (g), ciliated columnar cells (c), and tight junctions (tj) are presented. Basal cells (b) are located on the basal lamina (bl) adjacent to the lamina propia (lp) with blood vessels (v) (Martin et al., 1997).

In addition to passing through the mucus layer and penetration through the epithelium, substances may undergo metabolism by nasal enzymes present in the mucosa. Also, because of normal physiological mucocilliary clearance mechanisms, drugs have limited absorption time in the nasal cavity. Both of these are important factors to keep in mind for intranasal drug absorption. Furthermore, it has also been

suggested that redistribution of molecules within the nasal mucosa is a possibility (Newman et al., 1987). This recycling effect would be beneficial for topical agents; however, it may affect the absorption kinetics for drugs intended for systemic action.

Pharmacokinetic parameters following intranasal absorption will vary with administration of different agents. Values in the literature for time to reach maximum concentration in humans range from 5 min to 4 h (Illum and Davis, 1992; Hermens et al., 1992; Adjei et al., 1992; Hoffman and Ziv, 1997; Cincinelli et al., 1992; Eller et al., 1998; Hussain et al., 1980; Johnson et al., 1991). Also, bioavailability with intranasal delivery greatly varies and is dependent on numerous factors such as the drug itself, formulation differences, and the delivery system, all of which will affect nasal exposure, extent of absorption, and the pharmacokinetics of each particular agent or formulation (Fig. 2.5).

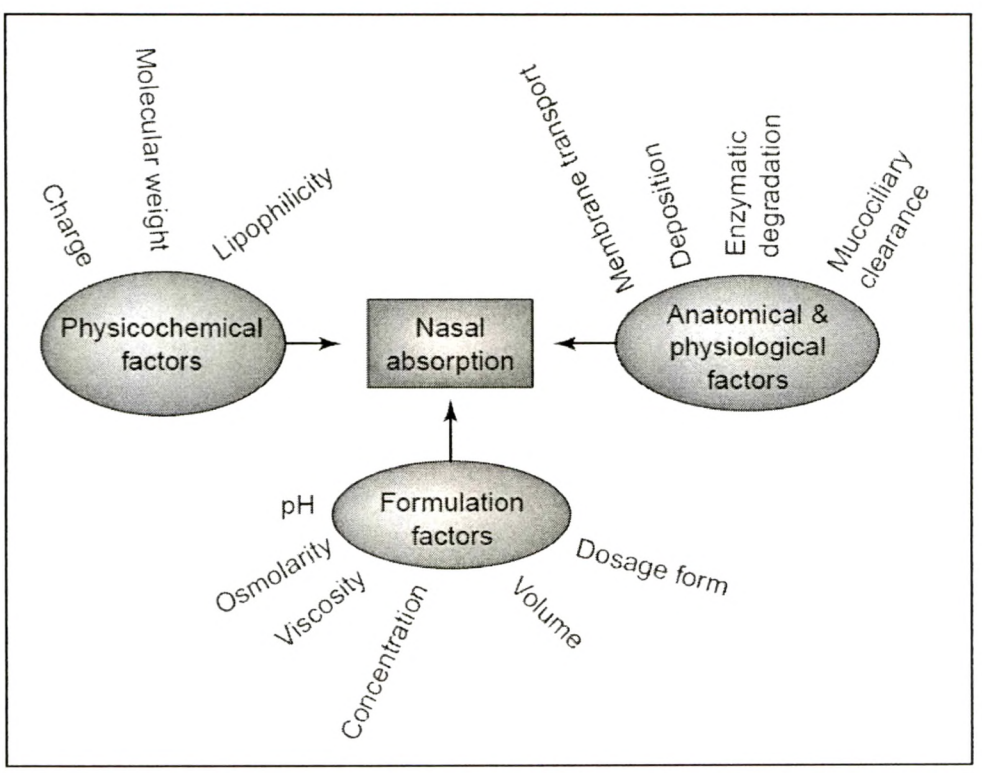


Fig. 2.5 The physicochemical, anatomical, physiological and formulation factors affecting the nasal absorption of drugs (Illum, 2002)

2.6 Nasal pathology with relevance to nasal drug absorption:

The nose may be affected by a number of pathological conditions. It is important to consider the effect that these may have on nasal drug absorption. The pH of the nasal fluid is normally around 5.5 - 6.5 but depends on air temperature, sleep, emotions, and food ingestion (Washington et al., 2000a). An increase in pH to 7 - 9 during acute and allergic rhinitis, rhinorrhoea, and chronic and acute sinusitis can be observed. Also diabetes mellitus has been shown to influence the nasal pH (Sachdeva et al., 1993).

Inhalation of cold, dry air can act as a physical stimulus inducing symptoms of rhinitis that are associated with an increase in osmolality from 280 - 290 to approximately 310 mosmol/kg (Togias et al., 1988). Stimulation of the nasal gland secretion with chili powder reduces the osmolality to approximately 238 mosmol/l, with simultaneous reduction of the sodium and potassium ion content of the nasal secretion (Knowles et al., 1997). Pathological conditions also affect the viscosity and viscoelasticity of the nasal mucus as well as the ciliary beat frequency (Atsuta and Majima, 1998; Majima et al., 1999). This variability in composition and properties of nasal fluid can greatly influence the performance of a nasally administered drug delivery system, especially when it relies on nasal fluid uptake for activation.

Bacteria, e.g. Haemophilus influenza and Staphylococcus epidermidis, are known to disturb normal synchronous ciliary motion, causing adjacent cilia to beat at different rates (Ferguson et al., 1988). Also disruption of epithelial cells with loss of a confluent epithelial field has been reported. Changes in ciliary structure occur in patients with long-standing allergic rhinosinusitis and variations in secreted mucus happen at times of acute allergen challenge (Maurizi et al., 1984).

Rhinorrhoea, a symptom in patients suffering from rhinosinusitis resulting from an allergic reaction or from infections such as the common cold, is often associated with increased nasal clearance, while nasal congestion, also a common symptom of rhinosinusitis, leads to a strongly reduced nasal clearance (Bond et al., 1984). However, no influence of rhinorrhoea on the nasal clearance of interferon was observed (Phillpotts et al., 1984). Increased mucociliary clearance can also be observed following acute exposure to tobacco smoke (Bascom et al., 1995). The pattern of nasal deposition from a spray device did not differ between normal subjects and those with nasal polyposis, although clearance was considerably slower in the

latter (Lee et al., 1984). Also allergic rhinitis, atrophic rhinitis and chronic sinusitis lead to a reduced nasal mucociliary clearance (Sakakura et al., 1983).

Patients with primary ciliary dyskinesia have no or dyskinetic beating cilia and thus a reduced mucociliary clearance, resulting in frequent infections of the respiratory system (Blouin et al., 2000). Cystic fibrosis patients exhibit also a reduced mucociliary clearance due to the abnormality of the mucus, while the cilia function is normal (Middleton et al., 1993). Mucociliary clearance in diabetes patients is also reduced compared to non-diabetic controls (Sachdeva et al., 1993).

2.7 Absorption Enhancers

Larger peptides and proteins are able to pass the nasal membrane using an endocytotic transport process but only in low amounts (Inagaki et al., 1985; Grass and Robinson, 1988). Nasal absorption of such polar drugs can be greatly improved by co-administration of absorption enhancing agents. Agents described in the literature for nasal drug delivery have included surfactants (laureth-9, sodium laurylsulfate), bile salts and bile salt derivatives (sodium glycocholate, sodium deoxycholate, sodium taurodihydrofusidate), fatty acids and fatty acid derivatives (linoleic acid), phospholipids (lysophosphatidylcholine, DDPC), various cyclodextrins (dimethyl-β-cyclodextrin, parenteral α-, β-, and γ-cyclodextrin), and cationic compounds (chitosan and derivatives, poly-L-arginine, poly-L-lysine) (De Ponti, 1991; Merkus et al., 1993; Illum, 1999, Natsume et al., 1999). These enhancers work by a variety of mechanisms but generally change the permeability of the epithelial cell layer by modifying the phospholipid bilayers, leaching of proteins from the membrane or even stripping off the outer layer of the mucosa. Some of these enhancers also have an effect on the tight junctions and / or work as enzymatic degradation inhibitors (Illum, 2003). With such absorption enhancing agents increased bioavailabilities were obtained, even for larger peptides such as insulin (Hinchcliffe and Illum, 1999). In animal studies it has been shown for a range of enhancing agents that there is a direct correlation between the absorption enhancing effect and the damage to the nasal mucosa (Illum, 1999). This is particularly true for bile salts and surfactants. For other enhancers, such as cyclodextrins and chitosan, the enhancing effect outweighs the damage caused to the mucosa. Hence, it is of great importance to consider the choice of absorption enhancer

for a nasally delivered drug that is not easily absorbed, especially in terms of potential nasal and systemic toxicity.

2.8 Considerations for Intranasal Product Design

2.8.1 Drug Formulation

Many factors play a role in the absorption of substances from the nasal cavity. These factors mostly center on the chemical attributes of a particular agent, the excipients in the formulation, and means of delivery. Molecular weight is believed to offer the best correlation with extent of absorption (Agarwal and Mishra, 1999; Fisher et al., 1987). Agents with a molecular weight less than 1000 are absorbed to a much larger degree than agents with molecular weights greater than 1000. For larger molecules, the addition of an adjuvant may be useful (McMartin et al., 1987). Not only is molecular weight important in absorption, but also the molecular size and shape of an agent. Cyclic-shaped molecules are better absorbed than those having a linear structure (McMartin et al., 1987). Particle size is also important because spray particles less than 10 μm may bypass the nasal cavity and be deposited in the lungs (Jones et al., 1997). For powder formulations, a particle size of greater than 50 μm has been shown to provide a favorable pattern of distribution in the nose (De Ascentus et al., 1996).

Systemic bioavailability decreases as the hydrophilicity of an agent increases (Corbo et al., 1989). Bioavailability is affected by pH and pKa factors as well (Ohwaki et al., 1985; Ohwaki et al., 1987; Behl et al., 1998; Shimoda et al., 1995). The pH of the drug, formulation, and nasal cavity all need to be taken into account. It is generally well known that un-ionized chemical species are capable of moving across membrane barriers more readily than ionized species, and thus, increased absorption usually occurs. However, drug formulations should not be extremely acidic because they may cause damage to the nasal epithelium and surface enzymes (Ohwaki et al., 1987; Shimoda et al., 1995). Although increased absorption could result from the structural damage, nasal integrity should not be compromised. Designing formulations so the final pH is within a range of 4.5–6.5 is suggested to minimize nasal irritation (Behl et al., 1998). Another formulation factor to consider is osmolarity. In one study, when the osmolarity of the solution was adjusted with mannitol, greater biological activity of the delivered agent was observed (Behl et al., 1998). Additionally, a study in rats demonstrated a solution of 0.462 M provided the best absorption of secretin although

shrinkage of epithelial cells was observed (Ohwaki et al., 1985). Other elements to consider when designing agents for intranasal administration are the solution concentration and volume to be delivered. One study focused on intranasal insulin delivery determined a higher concentration did not produce significantly greater activity (Valensi et al., 1996). On the other hand, the initial drug concentration has been found to influence nasal absorption (Chien et al., 1989; Hussain et al., 1985). Larger delivery volumes allow better distribution and coverage of the nasal passage, which theoretically would lead to greater drug absorption. One study suggests a larger volume with a weaker concentration is preferable to a small, very concentrated volume. Another study demonstrated higher plasma peak levels and biological response when the total spray volume delivered per nostril (100 μL) was delivered as two separate sprays (50 μL each) (Harris et al., 1988). The volume that can be reasonably delivered is also limited by the size of the nasal cavity. Behl et al. (Behl et al. 1998) suggest an upper limit of 25 mg/dose and a volume of 25–150 μL/nostril.

Since the normal physiological process of the nose, i.e., mucocilliary clearance, is critical to its defense mechanisms, disruption should be avoided. Substances delivered to the nasal cavity may cause inhibition of this system or enhance the clearance process (Schipper et al., 1991; van de Donk and Merkus, 1982). Enhancing this process in most cases would be undesirable because the time for drug absorption would decrease. So the potential nasal physiological effects a drug or formulation excipient may have must be considered as well. This should especially be evaluated for drugs intended for chronic therapy so prolonged mucosal dysfunction is avoided.

The discussion above has focused on formulation issues related to agents intended for systemic delivery. For local residence and effect in the nasal cavity there are other considerations. Of course, many of the items previously presented are important for either type of delivery. These include compatibility with the nasal tissue, for example, and appropriate pH and osmolarity, careful choice of a preservative if needed, and a reasonable concentration and volume for administration.

Additionally, drugs for local effect may be targeted to a specific area of the nasal cavity rather than general distribution. An example of this is treatment of nasal polyposis where the target area for drug disposition is the sinus mucosa (Denyer, 1999). Aqueous formulations are desirable for local delivery. If the drug molecule is

highly water-insoluble, it may be necessary to use wetting agents such as polysorbate 20 and sorbitan laurate to develop an aqueous suspension. Using this type of method, undissolved drug should be available at the local site of action (Denyer, 1999).

2.8.2 Dosage Form

Besides chemical and formulation issues, the dosage form is also important for intranasal administration. To date, delivery of substances to the nose has been via insufflation of powders, topical gels, sprays, drops, and nasal pledgets (Chien et al., 1989). Alternatively, powders appear to offer many benefits such as increased chemical stability, decreased need for preservative additions, administration of larger doses, and in some cases, greater bioavailability and absorption (De Ascentus et al., 1996). The choice of dosage form and the delivery system employed should be focused on feasibility and chemical stability, and should compliment the expected therapeutic use.

2.8.3 Delivery Device

The delivery device for a particular formulation should be based on several factors, including accuracy and dose reproducibility, cost, simple use for the patient, physiochemical characteristics of the drug and chosen dosage form, and protection from microbial contamination (Ugwoke et al., 2001). Metered-dose systems are considered the best for dose accuracy and reproducibility. Current delivery devices for liquid and powder intranasal formulations are the following (Ugwoke et al., 2001):

Liquid formulations - instillation catheter, dropper, unit-dose containers, squeeze bottle, pump spray, airless and preservative-free sprays, compressed air nebulizers, and metered-dose inhalers.

Powder formulations - insufflators, monodose inhalers, multidose inhalers, and pressurized metered-dose inhalers.

The delivery device and drug formulation should also be compatible with each other to prevent leaching and absorption, which could pose a risk for toxicity and may affect dose accuracy, respectively. Additionally, a device should be chosen on its ability to store the formulation appropriately without compromising stability (Ugwoke et al., 2001).

2.9 Mucoadhesive Drug Delivery Systems

Drugs for systemic medication are administered traditionally and routinely by oral and parenteral routes. Although generally convenient, both routes have a number of disadvantages. Oral administration results in the exposure of the drug to the harsh environment of the gastrointestinal tract and thus to potential chemical and enzymatic degradation (Langguth et al., 1997). After gastrointestinal absorption, the drug has to pass the liver, where, dependent on the nature of the drug, extensive first pass metabolism can take place with subsequent rapid clearance from the blood stream (Taki et al., 1998). Parenteral administration avoids drug degradation in the gastrointestinal tract and hepatic first pass clearance but due to pain or discomfort during injection, patient compliance is poor, particularly if multiple daily injections are required as e.g. in the insulin therapy (Hinchcliffe and Illum, 1999). Also injection related side effects like tissue necrosis and thrombophlebitis lead to low patient acceptability (Zhou, 1994). In addition, administration by injection requires trained personnel which add to the relatively high costs of parenteral medication.

Due to the clear advantages of accessibility, patient convenience, and permeability, nasal and pulmonary drug delivery are the most promising transmucosal delivery routes. The quantity of drug that can be delivered to the lung may be more limiting than that given nasally, but it is, of course, possible to give more than one dose. Davis (Davis, 1999) estimates that maximum doses of 30 mg and 50 mg active ingredient can be given nasally using solutions and powders, respectively, while the maximum pulmonary dose delivered by dry powder inhalers would be 5 times 3 mg. The final choice of the delivery route will depend on a variety of factors, but, in particular, on the nature of the drug, the dose of the active material, and the nature of treatment (acute vs. chronic). Any decision over choice will also need to consider patient convenience and cost.

Several mucosal routes have been investigated over the last decades as alternatives to oral and parenteral drug administration, including nasal, buccal, ocular, rectal, and vaginal mucosa (Ahuja et al., 1997). Their advantages are the easy accessibility and circumvention of the hepatic first pass metabolism. Mucosal bioavailability can vary between almost 100% for low molecular weight hydrophobic drugs (Hussain et al.,

1980) and below 1% for polar macromolecules (Zhou and Li Wan Po, 1991; Illum, 2003) depending on the nature of the delivered drug.

2.9.1 Mucoadhesion

When administering drugs to mucosal tissues, such as in the nasal cavity, it is helpful if the formulation stays in the right place long enough for the drug to be absorbed across the mucosa. It may, therefore, be beneficial to use a mucoadhesive agent in the formulation to achieve sufficient residence time.

Mucoadhesion, as the word suggests, refers to adhesion of matter to a mucus layer for an extended period of time (Gu et al., 1988). A mucoadhesive agent is thus a substance that adheres to mucus. The term bioadhesion is less specific and can be used to denote adhesion to any biological surface (Gu et al., 1988; Mathiowitz, E. and Chickering, 1999; Smart, 2005).

Mucoadhesive agents are usually polymers containing hydrogen bonding groups that can be used in wet formulations or, as in present study, in dry powders. The mechanisms behind mucoadhesion have not yet been fully elucidated, but a theory that has stuck is that close contact must first be established between the mucoadhesive agent and the mucus, followed by interpenetration of the mucoadhesive polymer and the mucin and finishing with the formation of entanglements and chemical bonds between the macromolecules (Mathiowitz, E. and Chickering, 1999). In the case of a dry polymer powder, the initial adhesion is most likely achieved by water movement from the mucosa into the formulation (Smart, 2005), which has also been shown to lead to dehydration and strengthening of the mucus layer (Mortazavi and Smart, 1993). The subsequent formation of van der Waals, hydrogen and, in the case of a positively charged polymer, electrostatic bonds between the mucin and the hydrated polymer promotes prolonged adhesion.

The mechanisms responsible for the formation of bonds are not yet completely clear. It is important to describe and understand the forces that are responsible for adhesive bond formation in order to develop mucoadhesive drug delivery systems. Most research has focused on analyzing mucoadhesive interactions between polymer hydrogels and soft tissue. The process involved in the formation of such mucoadhesive bonds has been described in three steps: first, wetting and swelling of

the polymer to permit intimate contact with the biological tissue, then interpenetration of mucoadhesive polymer chains and entanglement of polymer chains and mucin chains, and finally formation of weak chemical bonds between entangled chains (Duchene et al., 1988). It has been stated that at least one of the following **polymer characteristics** is required to obtain adhesion (Peppas and Buri, 1985):

(i) Sufficient quantities of hydrogen-bonding chemical groups (e.g. -OH and -COOH)
(ii) Anionic surface charges (also cationic polymers, e.g. chitosan, show bioadhesion)
(iii) High molecular weight
(iv) High chain flexibility
(v) Low surface tension that will induce spreading into the mucous layer.

Each of these characteristics favors the formation of bonds that are either chemical (e.g. ionic bonds, hydrogen bonds, van der Waals interactions) or mechanical (physical entanglement and / or interpenetration) in origin.

With respect to previous research for glues, adhesives and paints, five different **theories** have been adapted to the study of mucoadhesion (Mathiowitz and Chickering, 1999):

(i) The **electronic theory** is based on the assumption that the adhesive material and the target tissue have different electronic structures. When both surfaces come in contact, electron transfer occurs causing the formation of a double layer of electric charge at the interface. The bioadhesive force is believed to be due to attractive forces across the electrical double layer.

(ii) The **adsorption theory** states that the bioadhesive bond is formed due to van der Waals interactions, hydrogen bonds, and related forces. Although the individual forces are weak, the high number of interaction sites can produce intense adhesive strength. The adsorption theory is the most widely accepted theory of mucoadhesion.

(iii) The **wetting theory** was developed predominantly with regard to liquid adhesives. It uses interfacial tension to predict spreading and in turn adhesion.

(iv) The **diffusion theory** supports the concept that interpenetration and entanglement of mucoadhesive polymer chains and mucus chains produce semi permanent adhesive bonds. It is believed that the bond strength increases with the degree of the polymer

chain penetration into the mucus layer. Penetration of polymer chains into the mucus network, and vice versa, is dependent on the concentration gradients and the diffusion coefficients. Cross-linking of either component hinders the interpenetration but small chains and chain ends can still become entangled. For diffusion to occur, it is important to have good solubility of one component in the other. The mucoadhesive and the mucus should therefore be of similar structure.

(v) The **fracture theory** analyzes the forces required to separate two surfaces after adhesion.

It is therefore most applicable to studying mucoadhesion through mechanical measurements. When determining fracture properties of an adhesive union from separation experiments, failure of the adhesive bond must be assumed to occur at the mucoadhesive interface. However, it has been demonstrated that fracture rarely, if ever, happens at the interface but instead occurs close to the interface (Ponchel et al., 1987).

The largest groups of mucoadhesive materials are hydrophilic macromolecules containing numerous hydrogen bond-forming groups. These are called "wet" adhesives as they are activated by moistening. However, unless water uptake is restricted, they may overhydrate to form slippery mucilage (Smart, 1999). These hydrogel forming materials are nonspecific in action.

Drug delivery systems based on the concept of mucoadhesion have been widely investigated for various mucosal routes of administration including the nasal cavity. The prolonged residence time of the delivery system on the mucosa can result in higher drug absorption and consequently better bioavailability (Illum and Fisher, 1997).

A predicament with trying to increase the contact time by adhesion to mucus is that the residence time of the mucus itself is limited by mucociliary clearance. The normal transit time of a particle deposited on top of the nasal mucus layer is approximately 12-15 min (Schipper et al., 1991). However, dehydration of the mucus layer on contact with the mucoadhesive powder will increase the mucus viscosity and subsequently decelerate its clearance (Illum et al., 1987; Soane et al., 1999). Dry powder formulations should hence be especially well suited for nasal administration

as increased mucus viscosity would lower the normal requirements for the formation of secondary chemical bonds in order to prolong residence time.

With nasal, buccal, pulmonary, ocular, rectal, and vaginal mucosa as potential drug delivery sites, it is hard to identify the most suitable for clinical use. Only few studies were conducted to directly evaluate the different membrane permeabilities between these mucosal sites. Rojanaskul et al. (Rojanaskul et al., 1992) measured the electrical membrane resistance and the flux of the hydrophilic probe 6-carboxyfuorescein at various mucosal sites and found a good correlation between these two parameters. The data obtained was indicated that nasal and pulmonary epithelia are equally or only slightly less permeable than that of the intestine. The high permeability values of the respiratory tissue are a result of the presence of numerous aqueous pores through which water-soluble molecules can diffuse. Both large and small pores were reported in the nasal and pulmonary epithelium.

The nasal epithelium is leakier for peptide molecules than intestinal epithelia when using metabolically stable peptides as permeability tracers (McMartin et al., 1987). Opposite to other reports with mannitol and progesterone (Corbo et al., 1990) and 6-carboxyfluorescein (Rojanaskul et al., 1992), Aungst et al. (Aungst et al., 1988) demonstrated that nasal, buccal, and sublingual insulin administrations were less efficient than administration via rectal mucosa. This finding suggests that other factors like enzyme activity and absorptive surface area may play a role in determining the overall bioavailability. The large absorptive surface of the lung would make the pulmonary mucosa a very effective route of administration. This was also demonstrated by an absorption study in rats with different water-soluble compounds (phenol red, trypan blue, fluorescein isothiocyanate dextran molecular weight 4400 and 9100) which revealed bioavailabilities after mucosal administration of the order lung > small intestine > nasal cavity > large intestine > buccal cavity (Yamamoto et al., 2001). In the same study, the pharmacological availability of [ASU1,7]-eel calcitonin gave the order lung > nasal cavity > small intestine = large intestine ≥ buccal cavity which was attributed to the higher protease content in the small intestine compared to the nasal cavity. The proteolytic activity in different animals is relatively high in the rectal and ileal mucosa and comparatively low in the buccal, nasal, and vaginal mucosal tissue (Zhou and Li Wan Po, 1991).

2.10 Microparticles for Nasal Delivery

Mucoadhesive microparticles are used for prolonging the residence time in the nasal cavity and this concept was introduced by Illum et al. (Illum et al., 1987). Microspheres of albumin, starch, and DEAE-dextran absorbed water and formed a gel-like layer which was cleared slowly from the nasal cavity. Three hours after administration, 50% of the delivered amount of albumin and starch microspheres and 60% of the DEAE-dextran microspheres were still present at the site of deposition. It was suggested that an increased contact time could increase the absorption efficiency of drugs. As proposed, the relative intranasal bioavailability (vs. s.c.) of human growth hormone in sheep was increased from 0.1% for the solution to 2.7% for the degradable starch microsphere formulation (Illum et al., 1990). Addition of the absorption enhancer lysophosphatidylcholine further enhanced the growth hormone absorption (relative bioavailability 14.4%). Björk and Edman (Björk and Edman, 1990) showed that plasma glucose reduction after nasal insulin administration was comparable for degradable starch microspheres (cross-linked with epichlorohydrin) and insoluble starch powder (molecular weight 25 kDa) but significantly lower for soluble starch powder (molecular weight 11 kDa). It was therefore concluded that crucial parameters for the absorption promoting effect of microspheres are water absorption and water insolubility. No alteration of the nasal mucosa was observable by scanning electron microscopy after 8 weeks of twice daily administration of starch microspheres, except slight hyperplasia in the septum wall (Edman et al., 1992).

Although DEAE-dextran microspheres were retained strongly in the nasal cavity (Illum et al., 1987), they were not successful in promoting nasal insulin absorption in rats (Rydén and Edman, 1992). The insulin was too intensely bound to the DEAE-groups to be released by a solution with an ionic strength corresponding to physiological conditions. Dextran microparticles without ion exchange groups induced a 25% decrease in blood glucose level about 40 min after administration compared to initial levels. In a later study, dextran microspheres with different insulin distribution were compared (Pereswetoff-Morath and Edman, 1995). When insulin was situated on the microsphere surface, a 52% reduction in plasma glucose was induced 30 min after administration in rats. However, microspheres, which included insulin in the sphere matrix, reached a maximum plasma glucose level reduction of

30% after 60 min. Possibly the limited amount of fluid in the nasal cavity is responsible for the differences observed, because the microspheres needed to be completely swollen to release the entire amount of incorporated insulin (Pereswetoff-Morath, 1998).

Chitosan has also a high potential as particulate drug delivery carrier. In vivo studies in sheep showed a half-life of nasal clearance for chitosan microparticles of 115 min compared to 43 min for a solution of the polymer (Soane et al., 2001). In general, chitosan powder formulations, whether in form of microparticles or powders, were shown to provide a better absorption enhancing effect than chitosan solutions (Illum, 2003). Hence, a chitosan microsphere formulation for a LHRH analogue provided an absolute bioavailability in the order of 40% in sheep (Illum et al., 2000b). Numerous patents were developed based on chitosan microparticles for nasal drug delivery (Watts and Illum, 1996; Watts and Illum, 1998b; Watts and Illum, 2001). Chitosan nanoparticles were also exploited as nasal drug delivery system. Insulin loaded chitosan nanoparticles enhanced the nasal absorption of insulin to a greater extent than chitosan solution (Fernandez-Urrusuno et al., 1999). The amount and molecular weight range of chitosan investigated did not have a significant effect on insulin response.

Among other microparticulate delivery devices, hyaluronic acid ester microspheres were successfully examined as potential drug delivery system for insulin reaching a relative bioavailability (vs. s.c.) of 11% in sheep (Illum et al., 1994a). Bioadhesive microparticles of pH-sensitive co-polymers of polymethacrylic acid and polyethylene glycol were investigated as potential nasal drug delivery system for budesonide (Nakamura et al., 1999).

The mechanism by which dry-powder / particulate carriers enhance the nasal bioavailability of drugs is only incompletely understood. Several hypotheses have been presented, which are usually quite specific for the physicochemical parameters of the respective delivery system, and also combinations of these proposed **mechanisms** seem likely:

(i) increased nasal contact time due to mucoadhesion for starch microspheres (Illum et al., 1987),

(ii) increased nasal residence time due to the insolubility of the particulate carrier, e.g. for microcrystalline cellulose and calcium phosphate (Nagai et al., 1984; Ishikawa et al., 2002),

(iii) particulate uptake via macrophages for PLA-microparticles (Almeida et al., 1993),

(iv) opening of tight junctions by dehydration of mucosal cells due to the connection to the cell cytoskeleton, e.g. in the case of degradable starch microspheres (Björk et al., 1995),

(v) opening of tight junctions by binding of Ca^{2+} for cross-linked dextran microspheres, alginic acid, and microcrystalline cellulose. Oechslein et al. (Oechslein et al., 1996) found a better correlation between the Ca^{2+}-binding capacity of various powder excipients and the bioavailability of octreotide in rats than with their water uptake behavior. Although not described for a nasal powder formulation, thiolated polymers also increased the absorption of drugs by opening of tight junctions but by a different mechanism (Clausen et al., 2002). Here, the reducing activity of the thiolated polymer (polycarbophil-cystein conjugate) on oxidized glutathione led to an increased glutathione level in the mucosa. Glutathione inhibited the enzyme tyrosine phosphatase, which in turn is responsible for the dephosphorylation of the tyrosine residues of occludin, a major tight junction protein. This dephosphorylation of occludin then resulted in opening of the junctions and thus increased paracellular transport.

Although particulate / powder formulations for nasal administration offer numerous advantages with respect to retention in the nasal cavity, absorption enhancement, and stability due to the absence of water and lack of preservatives, the delivery of these powder dosage forms to the nose is complex and so far no product has entered the market. Nasal powders require sophisticated delivery devices and formulation optimization. Parameters such as particle size and shape, density, and flow characteristics influence the distribution in the nose (Kublik and Vidgren, 1998). Insufflators (mainly for study purposes), mono-dose and multidose dry powder inhalers (e.g. Miat, Milano, Italy; Pfeiffer, Randolfzell, Germany; Valois, Le Neubourg, France; Orion, Helsinki, Finland; Teijin, Osaka, Japan) have been developed for the nasal application. Dry powder inhalers ensure correct dosing and achieve better patient compliance. A new development enables the application of freeze-dried powder, which can be lyophilized directly in the device.

2.11 DRUG PROFILES

2.11.1 CARVEDILOL

(Martindale, 2005; Rang et al., 2003; Weir and Darjie, 2005)

Category: Non-cardioselective beta blocker.

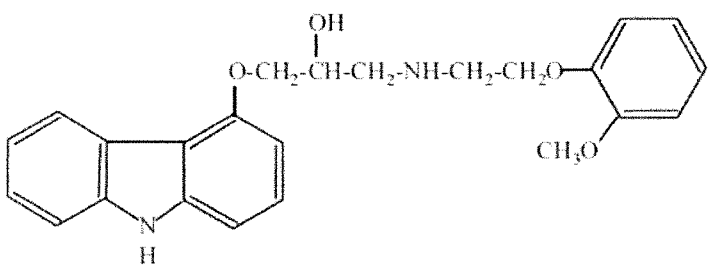
CAS Number: *72956–09–3*

Proprietary names: Coreg, Carvipress; Dilatrend; Dimitone; Eucardic; Kredex; Querto.

Molecular formula: $C_{24}H_{26}N_2O_4$

Molecular Weight: 406.5

Structural Formula and Chemical Name:



(±) -1-(9 H-carbazol-4-yloxy)-3-{[2-(2-methoxyphenoxy) ethyl] amino} propan-2-ol.

Physicochemical Properties:

Appearance and colour: A white or almost white crystalline powder.

Solubility: It is practically insoluble in water; freely soluble in dimethyl sulfoxide; soluble in methylene chloride and in methanol; sparingly soluble in ethanol and in isopropyl alcohol; slightly soluble in ethyl ether.

Partition Coefficient. Log *P*(octanol/water), 4.19.

Melting point: 114° to 115°.

Mechanism of Action:

The mechanism for the beneficial effects of Carvedilol in congestive heart failure has not been established. Possible mechanisms include neurohormonal inhibition, β-blockade, balanced vasodilation (reduced preload and after load), antioxidant activity, potent antiischaemic activity, and inhibition of neutrophil adhesion. Antioxidant activity and inhibition of neutrophil adhesion have been demonstrated in in-vitro and in vivo animal models.

Carvedilol reduces the peripheral vascular resistance by vasodilation predominantly mediated through selective alpha1 - antagonism and beta blockade prevents reflex tachycardia with the net result that heart rate is slightly decreased.

Pharmacokinetics:

Absorption:

Carvedilol is rapidly and extensively absorbed following oral administration. The absolute bioavailability of Carvedilol is approximately 25%. Plasma levels peak approximately 1 hour after an oral dose. Carvedilol undergoes stereoselective first-pass metabolism with plasma levels of R (+)-Carvedilol approximately 2 to 4-fold higher than S (-)-Carvedilol following oral administration in healthy subjects. Plasma levels increase in a dose-proportional manner.

Distribution:

Carvedilol is highly lipophilic, therefore it is 98% bound to plasma proteins, primarily albumin. The volume of distribution is approximately 2 L/kg and is increased in patients with liver disease. When used as directed, Carvedilol is unlikely to accumulate during long term treatment.

Metabolism:

In all animal species studies, and also in humans, Carvedilol is extensively metabolised into a variety of metabolites which are mainly excreted in the bile. The first-pass effect after oral administration amounts to about 60-75%; enterohepatic circulation of Carvedilol and/or its metabolites has been shown in animals. The major P450 enzymes responsible for the metabolism of both R(+) and S(-) Carvedilol in human liver microsomes were identified as CYP2D6 and CYP2C9, and to a lesser extent CYP3A4, CYP2C19 and CYP2E1.

Elimination:

After oral administration, the elimination half-life of Carvedilol is approximately 6 to 10 hours. Plasma clearance ranges from 500 to 700 mL/min. Elimination is mainly biliary, with the primary route of excretion being via the faeces. A minor portion is eliminated via the kidneys. The pharmacokinetics of Carvedilol is affected by age.

Usage and Administration:

Carvedilol is used in the management of hypertension and angina pectoris, and as an adjunct to standard therapy in symptomatic heart failure. It is also used to reduce mortality in patients with left ventricular dysfunction following myocardial infarction.

In hypertension, Carvedilol is given at an initial dose of 12.5 mg once daily by mouth, increased after two days to 25 mg once daily. Alternatively, an initial dose of 6.25 mg is given twice daily, increased after one to two weeks to 12.5 mg twice daily. The dose may be increased further, if necessary, at intervals of at least two weeks, to 50 mg once daily or in divided doses. A dose of 6.25/12.5 mg once daily may be adequate for elderly patients.

In angina pectoris, an initial dose of 12.5 mg is given twice daily by mouth, increased after two days to 25 mg twice daily.

In heart failure, the initial dose is 3.125 mg twice daily by mouth. It should be taken with food to reduce the risk of hypotension. If tolerated, the dose should be doubled after two weeks to 6.25 mg twice daily and then increased gradually, at intervals of not less than two weeks, to the maximum dose tolerated; this should not exceed 25 mg twice daily in patients with severe heart failure or in those weighing less than 85 kg, or 50 mg twice daily in patients with mild to moderate heart failure weighing more than 85 kg.

In patients with left ventricular dysfunction following myocardial infarction, the initial dose is 6.25 mg twice daily, increased after 3 to 10 days, if tolerated, to 12.5 mg twice daily and then to a target dose of 25 mg twice daily. A lower initial dose may be used in symptomatic patients.

Adverse Effects:

Hypotension is the common effect observed with the patients taking Carvedilol. Acute renal failure and renal abnormalities have been reported in patients with heart failure. Pruritus and elevated serum transaminase concentrations occurred.

Contraindications:

Carvedilol is contraindicated in patients with NYHA class IV decompensated cardiac failure requiring intravenous inotropic therapy, bronchial asthma or related bronchospastic conditions, second-or third-degree AV block, sick sinus syndrome (unless a permanent pacemaker is in place), cardiogenic shock or severe bradycardia. Use of Carvedilol in patients with clinically manifest hepatic impairment is not recommended. Carvedilol is contraindicated in patients with hypersensitivity to the drug.

Formulations Available:

Tablets: White, oval film-coated tablets

3.125 mg in bottles of 100; 6.25 mg in bottles of 100; 12.5 mg in bottles of 100; 25 mg in bottles of 100.

Analytical Techniques:

Colorimetric estimation:

A simple, accurate and sensitive spectrophotometric method for the estimation of Carvedilol in pharmaceuticals is developed by Ajoy and Dilipkumar (Ajoy and Dilipkumar, 2001). The method is based on the formation of a yellow colour with dilute HCl and sodium nitrite. This colour showed absorption maxima at 400 nm and obeys Beer's law up to 20 g/ml. The colour was found to be stable for 1 to 3 hr.

Ultraviolet Spectrum:

Aqueous acid (2.0 M HCl)—241, 285, 320, 332 nm; (methanol)—224, 243, 286, 319, 332 nm (Moffat et al., 2004).

High Performance Liquid Chromatography:

System HZ—retention time carvedilol, 5.2 min; desmethylcarvedilol, 4.4 min.

Column: Spherisorb octyl (125 × 5 mm i.d., 5 μm). Mobile phase: acetonitrile:water (45:55) containing dibutylamine (5 mM), 2 mL/min flow rate. Fluorescence detection (λ_{ex}=40 nm, λ_{em}=340 nm). Retention time: 3.8 min. (Varin et al., 1986).

2.11.2 NITRENDIPINE

(Martindale, 2005; Rang et al., 2003)

Category: Pyridine calcium channel blocker.

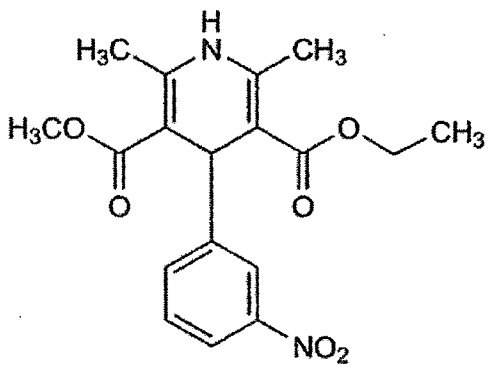
CAS Number: 39562-70-4

Proprietary names: Bayotensin; Baypresol; Baypress; Bylotensin; Deiten; Gericin; Monopress; Nidrel; Niprina; Nitrendepat; Nitrepress; Subtension; Tensogradal; Trendinol; Vastensium.

Molecular formula: $C_{18}H_{20}N_2O_6$

Molecular Weight: 360.4

Structural Formula and Chemical Name:



methyl ethyl 2, 6-dimethyl-4-(3-nitrophenyl)-1, 4-dihydropyridine-3, 5-dicarboxylate; 1, 4-Dihydro-2, 6-dimethyl-4-(meta-nitrophenyl)-3, 5-pyridinedicarboxylic acid, ethyl methyl ester; ethyl methyl 1, 4-dihydro-2,6-dimethyl-4-(meta-nitrophenyl)-3,5-pyridine dicarboxylate; 3-ethyl-5methyl-1, 4-dihydro-2, 6-dimethyl-4-(3-nitrophenyl) 3, 5-pyridine dicarboxylate.

Physicochemical Properties:

Appearance and colour: A yellow crystalline powder.

Solubility: It is practically insoluble in water; sparingly soluble in absolute alcohol and methanol; freely soluble in ethyl acetate. Exposure to light leads to formation of nitrophenylpyridine derivatives. Soluble in DMSO (25 mg/ml).

Partition Coefficient: Log *P*(octanol/water), 2.88.

Melting point: 156-160°C.

Mechanism of Action:

Nitrendipine, a dihydropyridine calcium-channel blocker, is used alone or with an angiotensin-converting enzyme inhibitor, to treat hypertension, chronic stable angina pectoris, and Prinzmetal's variant angina. Nitrendipine is similar to other peripheral vasodilators. Nitrendipine inhibits the influx of extra cellular calcium across the myocardial and vascular smooth muscle cell membranes possibly by deforming the channel, inhibiting ion-control gating mechanisms, and/or interfering with the release of calcium from the sarcoplasmic reticulum. The decrease in intracellular calcium inhibits the contractile processes of the myocardial smooth muscle cells, causing dilation of the coronary and systemic arteries, increased oxygen delivery to the myocardial tissue, decreased total peripheral resistance, decreased systemic blood pressure, and decreased after load.

Pharmacokinetics:

It is well absorbed orally from the gastrointestinal tract, metabolized in the liver and undergoes extensive first pass metabolism by cytochrome P450 enzyme CYP3A4. It has very poor absolute bioavailability (10–20%). A very high degree of variability of pharmacokinetic parameters is observed due to the differences in hepatic metabolism and plasma protein concentration (Soons et al., 1991; Reynolds, 1996). It is excreted via urine (as metabolites) and via faeces (remaining dose).

Usage and Administration:

Hypertension: ***Adult:*** 20 mg daily as a single dose or in 2 divided doses; may be increased to 20 mg bid as necessary to control resistant hypertension.

Elderly: Initially, 10 mg daily.

Contraindications: It is contraindicated in Hypersensitivity reaction, Acute MI, Hypotension, Lactation, Cardiogenic shock and in severe aortic stenosis.

Formulations Available:

Tablets: Cardif (Concept), Nitrepin (USV). 10 mg, 20 mg

Analytical Techniques:

Ultraviolet Spectrum:

Aqueous acid—236, 203, 350 nm.

High Performance Liquid Chromatography.

System HX—RI 625; system HY—RI 554; system HAA—retention time 22.1 min.

Column: Lichrocart (125 × 4.0 mm i.d., 5 μm). Mobile phase: methanol:water:acetonitrile (45:45:10), flow rate 1.2 mL/min. UV detection (λ=235 nm). Retention time: 5.94 min.

Column: ODS (Jasco, 250 × 4.0 mm i.d., 5 μm). Mobile phase: acetonitrile:potassium dihydrogen phosphate (10 mM, pH 4.5), flow rate 1.5 mL/min. Internal standard: felodipine. UV detection (λ=250 nm). Retention time(s): nitrendipine, 6.93 min; I. S., 12.20 min (Moffat et al., 2004).

2.12 EXCIPIENTS

2.12.1 Chitosan

Chitosan occurs as odorless, white or creamy-white powder or flakes. Fiber formation is quite common during precipitation and the chitosan may look 'cottonlike'.

Synonyms

2-Amino-2-deoxy-(1,4)-β-D-glucopyranan; deacetylated chitin; deacetylchitin; β-1,4-poly-Dglucosamine; poly-D-glucosamine; poly-(1,4-β-D-glucopyranosamine).

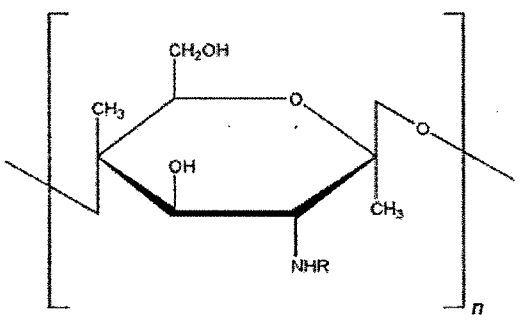
Chemical Name and CAS Registry Number

Poly-β-(1, 4)-2-Amino-2-deoxy-D-glucose [9012-76-4]

Empirical Formula and Molecular Weight

Partial deacetylation of chitin results in the production of chitosan, which is a polysaccharide comprising copolymers of glucosamine and *N*-acetylglucosamine. Chitosan is the term applied to deacetylated chitins in various stages of deacetylation and depolymerization and it is therefore not easily defined in terms of its exact chemical composition. A clear nomenclature with respect to the different degrees of *N*-deacetylation between chitin and chitosan has not been defined, (Kumar, 2000) and as such chitosan is not one chemical entity but varies in composition depending on the manufacturer. In essence, chitosan is chitin sufficiently deacetylated to form soluble amine salts. The degree of deacetylation necessary to obtain a soluble product must be greater than 80–85%. Chitosan is commercially available in several types and grades that vary in molecular weight by 10 000–1 000 000, and vary in degree of deacetylation and viscosity (Genta et al., 1998).

Structural Formula



R = H or $COCH_3$

Functional Category

Coating agent; disintegrant; film-forming agent; mucoadhesive; tablet binder; viscosity increasing agent.

Pharmacopoeial Specifications

Table 2.2 Pharmacopoeial specifications for chitosan.

Test	PhEur 2005
Identification	+
Characters	+
Appearance of solution	+
Matter insoluble in water	≤0.50%
pH (1% w/v solution)	4.0–6.0
Viscosity	+
Degree of deacetylation	+
Chlorides	10.0–20.0%
Heavy metals	≤40 ppm
Loss on drying	≤10%
Sulfated ash	≤1.00%

Typical Properties

Chitosan is a cationic polyamine with a high charge density at pH <6.5; and so adheres to negatively charged surfaces and chelates metal ions. It is a linear polyelectrolyte with reactive hydroxyl and amino groups (available for chemical reaction and salt formation) (Singla and Chawla, 2001). The properties of chitosan relate to its polyelectrolyte and polymeric carbohydrate character. The presence of a number of amino groups allows chitosan to react chemically with anionic systems, which results in alteration of physicochemical characteristics of such combinations.

The nitrogen in chitosan is mostly in the form of primary aliphatic amino groups. Chitosan therefore undergoes reactions typical of amines: for example, *N*-acylation and Schiff reactions (Kumar, 2000). Almost all functional properties of chitosan depend on the chain length, charge density, and charge distribution (Dodane and Vilivalam, 1998). Numerous studies have demonstrated that the salt form, molecular weight, and degree of deacetylation as well as pH at which the chitosan is used all influence how this polymer is utilized in pharmaceutical applications (Singla and Chawla M, 2001).

Acidity/alkalinity: pH = 4.0–6.0 (1% w/v aqueous solution)

Density: 1.35–1.40 g/cm^3

Glass transition temperature: 203°C (Sakurai et al., 2000)

Moisture content: Chitosan adsorbs moisture from the atmosphere, the amount of water adsorbed depending upon the initial moisture content and the temperature and relative humidity of the surrounding air (Gocho et al., 2000).

Particle size distribution:< 30 μm

Solubility: Sparingly soluble in water; practically insoluble in ethanol (95%), other organic solvents, and neutral or alkali solutions at pH above approximately 6.5. Chitosan dissolves readily in dilute and concentrated solutions of most organic acids and to some extent in mineral inorganic acids (except phosphoric and sulfuric acids). Upon dissolution, amine groups of the polymer become protonated, resulting in a positively charged polysaccharide (RNH^{+}_{3}) and chitosan salts (chloride, glutamate, etc.) that are soluble in water; the solubility is affected by the degree of deacetylation (Singla and Chawla, 2001). Solubility is also greatly influenced by the addition of salt to the solution. The higher the ionic strength, the lower the solubility as a result of a salting-out effect, which leads to the precipitation of chitosan in solution (Errington et al., 1993). When chitosan is in solution, the repulsions between the deacetylated units and their neighboring glucosamine units cause it to exist in an extended conformation. Addition of an electrolyte reduces this effect and the molecule possesses a more random, coil-like conformation (Skaugrud, 1991).

Viscosity (dynamic): A wide range of viscosity types is commercially available. Owing to its high molecular weight and linear, unbranched structure, chitosan is an excellent viscosity-enhancing agent in an acidic environment. It acts as a pseudo-plastic material, exhibiting a decrease in viscosity with increasing rates of shear (Singla and Chawla, 2001). The viscosity of chitosan solutions increases with increasing chitosan concentration, decreasing temperature, and increasing degree of deacetylation (Table 2.3) (Skaugrud, 1991).

Table 2.3 Typical viscosity (dynamic) values for chitosan 1% w/v solutions in different acids (Skaugrud, 1991)

Acid	1% acid concentration		5% acid concentration		10% acid concentration	
	Viscosity (mPa s)	pH	Viscosity (mPa s)	pH	Viscosity (mPa s)	pH
Acetic	260	4.1	260	3.3	260	2.9
Adipic	190	4.1	—	—	—	—
Citric	35	3.0	195	2.3	215	2.0
Formic	240	2.6	185	2.0	185	1.7
Lactic	235	3.3	235	2.7	270	2.1
Malic	180	3.3	205	2.3	220	2.1
Malonic	195	2.5	—	—	—	—
Oxalic	12	1.8	100	1.1	100	0.8
Tartaric	52	2.8	135	2.0	160	1.7

Stability and Storage Conditions

Chitosan powder is a stable material at room temperature, although it is hygroscopic after drying. Chitosan should be stored in a tightly closed container in a cool, dry place. The PhEur 2005 specifies that chitosan should be stored at a temperature of 2–8°C.

Incompatibilities

Chitosan is incompatible with strong oxidizing agents.

Method of Manufacture

Chitosan is manufactured commercially by chemically treating the shells of crustaceans such as shrimps and crabs. The basic manufacturing process involves the removal of proteins by treatment with alkali and of minerals such as calcium carbonate and calcium phosphate by treatment with acid (Kumar, 2000; Skaugrud, 1991). Before these treatments, the shells are ground to make them more accessible. The shells are initially deproteinized by treatment with an aqueous sodium hydroxide 3–5% solution. The resulting product is neutralized and calcium is removed by treatment with an aqueous hydrochloric acid 3–5% solution at room temperature to precipitate chitin. The chitin is dried so that it can be stored as a stable intermediate for deacetylation to chitosan at a later stage. *N*-deacetylation of chitin is achieved by

treatment with an aqueous sodium hydroxide 40–45% solution at elevated temperature (110°C), and the precipitate is washed with water. The crude sample is dissolved in acetic acid 2% and the insoluble material is removed. The resulting clear supernatant solution is neutralized with aqueous sodium hydroxide solution to give a purified white precipitate of chitosan. The product can then be further purified and ground to a fine uniform powder or granules. The animals from which chitosan is derived must fulfil the requirements for the health of animals suitable for human consumption to the satisfaction of the competent authority. The method of production must consider inactivation or removal of any contamination by viruses or other infectious agents.

Applications in Pharmaceutical Formulation or Technology

Chitosan is used in cosmetics and is under investigation for use in a number of pharmaceutical formulations. The suitability and performance of chitosan as a component of pharmaceutical formulations for drug delivery applications has been investigated in numerous studies (Kumar, 2000; Illum, 1998; Paul and Sharma; Singla and Chawla, 2001; Dodane and Vilivalam, 1998). These include controlled drug delivery applications (Nakatsuka and Andrady, 1992; Li et al., 1992), use as a component of mucoadhesive dosage forms (He and Davis, 1998; He and Davis, 1999), rapid release dosage forms (Sawayangi et al., 1982; Shirashi et al., 1990), improved peptide delivery (Leussen et al., 1994; Leussen et al., 1997), colonic drug delivery systems (Tozaki et al., 1999a; Tozaki et al., 1999b), and use for gene delivery (Leong et al., 1998). Chitosan has been processed into several pharmaceutical forms including gels (Kristl et al, 1993; Tasker et al., 1997), films (Remunan-Lopez et al., 1998; Senel et al., 2000), beads (Kofuji et al., 1999; Sezer and Akbuga, 1999), microspheres (Ganza-Gonzalez et al., 1999; Huang et al., 1999), tablets (Sabnis et al., 1997), and coatings for liposomes (Takeuchi et al., 1996). Furthermore, chitosan may be processed into drug delivery systems using several techniques including spray drying (He et al., 1999), coacervation (Bayomi et al., 1998), and conventional granulation processes (Miyazaki et al., 1995).

Safety

Chitosan is being investigated widely for use as an excipient in oral and other pharmaceutical formulations. It is also used in cosmetics. Chitosan is generally regarded as a nontoxic and nonirritant material. It is biocompatible with both healthy

and infected skin (Gooday et al., 1986). Chitosan has been shown to be biodegradable (Gebelein and Dunn, 1990; Kumar, 2000).

LD_{50} (mouse, oral): >16 g/kg (Arai et al., 1968)

Handling Precautions

Observe normal precautions appropriate to the circumstances and quantity of material handled. Chitosan is combustible; open flames should be avoided. Chitosan is temperature sensitive and should not be heated above 200°C. Airborne chitosan dust may explode in the presence of a source of ignition, depending on its moisture content and particle size. Water, dry chemicals, carbon dioxide, sand, or foam fire-fighting media should be used.

Chitosan may cause skin or eye irritation. It may be harmful if absorbed through the skin or if inhaled and may be irritating to mucous membranes and the upper respiratory tract. Eye and skin protection and protective clothing are recommended; wash thoroughly after handling.

Prolonged or repeated exposure (inhalation) should be avoided by handling in a well ventilated area and wearing a respirator.

Regulatory Status

Chitosan is registered as a food supplement in some countries.

2.12.2 Sodium Alginate

Sodium alginate occurs as an odorless and tasteless, white to pale yellowish-brown colored powder.

Synonyms

Algin; alginic acid, sodium salt; E401; *Kelcosol*; *Keltone*; *Protanal*; sodium polymannuronate.

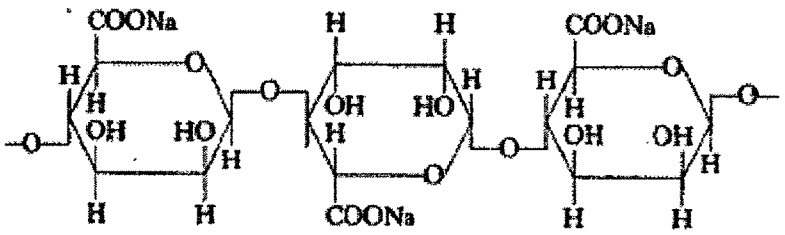
Chemical Name and CAS Registry Number

Sodium alginate [9005-38-3]

Empirical Formula and Molecular Weight

Sodium alginate consists chiefly of the sodium salt of alginic acid, which is a mixture of polyuronic acids composed of residues of D-mannuronic acid and L-guluronic acid. The block structure and molecular weight of sodium alginate samples has been investigated (Johnson et al., 1997).

Structural Formula



Functional Category

Stabilizing agent; suspending agent; tablet and capsule disintegrant; tablet binder; viscosityincreasing agent.

Pharmacopeial Specifications

Table 2.4 Pharmacopeial specifications for sodium alginate

Test	PhEur 2005	USPNF 23
Characters	+	+
Identification	+	+
Appearance of solution	+	+
Microbial limits	≤1000/g	≤200/g
Loss on drying	≤15.00%	≤15.00%
Ash	-	18.0–27.0%
Sulfated ash	30.0–36.0%	-
Arsenic	-	≤1.5 ppm
Calcium	≤1.50%	-
Chlorides	≤1.00%	-
Lead	-	≤0.001%
Heavy metals	≤20 ppm	≤0.004%
Assay (dried basis)	-	90.8–106.0%

Typical Properties

Acidity/alkalinity: pH ≈7.2 for a 1% w/v aqueous solution.

Solubility: Practically insoluble in ethanol (95%), ether, chloroform, and ethanol/water mixtures in which the ethanol content is greater than 30%. Also, practically insoluble in other organic solvents and aqueous acidic solutions in which the pH is less than 3. Slowly soluble in water, forming a viscous colloidal solution.

Viscosity (dynamic): Various grades of sodium alginate are commercially available that yield aqueous solutions of varying viscosity. Typically, a 1% w/v aqueous solution, at 20°C, will have a viscosity of 20–400 mPa s (20–400 cP). Viscosity may vary depending upon concentration, pH, temperature, or the presence of metal ions (Bugaj et al., 1996). Above pH 10, viscosity decreases.

Stability and Storage Conditions

Sodium alginate is a hygroscopic material, although it is stable if stored at low relative humidities and a cool temperature. Aqueous solutions of sodium alginate are most stable at pH 4–10. Below pH 3, alginic acid is precipitated. A 1% w/v aqueous solution of sodium alginate exposed to differing temperatures had a viscosity 60–80% of its original value after storage for 2 years (Pávics, 1970). Solutions should not be stored in metal containers. Sodium alginate solutions are susceptible on storage to microbial spoilage, which may affect solution viscosity. Solutions are ideally sterilized using ethylene oxide, although filtration using a 0.45 μm filter also has only a slight adverse effect on solution viscosity (Coates and Richardson, 1974). Heating sodium alginate solutions to temperatures above 70°C causes depolymerization with a subsequent loss of viscosity. Autoclaving of solutions can cause a decrease in viscosity, which may vary depending upon the nature of any other substances present (Vandenbossche and Remon, 1993). Gamma irradiation should not be used to sterilize sodium alginate solutions since this process severely reduces solution viscosity (Hartman et al., 1975).

Preparations for external use may be preserved by the addition of 0.1% chlorocresol, 0.1% chloroxylenol, or parabens. If the medium is acidic, benzoic acid may also be used. The bulk material should be stored in an airtight container in a cool, dry place.

Incompatibilities

Sodium alginate is incompatible with acridine derivatives, crystal violet, phenyl mercuric acetate and nitrate, calcium salts, heavy metals, and ethanol in concentrations greater than 5%. Low concentrations of electrolytes cause an increase in viscosity but high electrolyte concentrations cause salting-out of sodium alginate; salting-out occurs if more than 4% of sodium chloride is present.

Method of Manufacture

Alginic acid is extracted from brown seaweed and is neutralized with sodium bicarbonate to form sodium alginate.

Applications in Pharmaceutical Formulation or Technology

Sodium alginate is used in a variety of oral and topical pharmaceutical formulations (Tonnesen and Karlsen, 2002). In tablet formulations, sodium alginate may be used as both a binder and disintegrants (Sakr et al., 1978); it has been used as a diluent in capsule formulations (Veski and Marvola, 1993). Sodium alginate has also been used in the preparation of sustained-release oral formulations since it can delay the dissolution of a drug from tablets (Holte et al., 2003; Azarmi et al., 2003), capsules (Veski et al., 1994), and aqueous suspensions (Zatz et al., 1987).

In topical formulations, sodium alginate is widely used as a thickening and suspending agent in a variety of pastes, creams, and gels, and as a stabilizing agent for oil-in-water emulsions. Recently, sodium alginate has been used for the aqueous microencapsulation of drugs (Bodmeier and Wang, 1993), in contrast with the more conventional microencapsulation techniques which use organic solvent systems. It has also been used in the formation of nanoparticles (Rajaonarivony et al., 1993).

The adhesiveness of hydrogels prepared from sodium alginate has been investigated (Vennat et al., 1998) and drug release from oral mucosal adhesive tablets (Miyazaki et al., 1995), and buccal gels (Attia et al., 2004; Mohammed and Kheder, 2003), based on sodium alginate have been reported. Other novel delivery systems containing sodium alginate include ophthalmic solutions that form a gel *in situ* when administered to the eye (Cohen et al., 1997; Balasubramaniam and Pandit, 2003); an *in situ* forming gel containing paracetamol for oral administration (Kubo et al., 2003); and a freeze-dried device intended for the delivery of bone-growth factors (Duggirala and DeLuca, 1996).

Hydrogel systems containing alginates have also been investigated for delivery of proteins and peptides (Gombotz and Pettit, 1995).

Therapeutically, sodium alginate has been used in combination with an H_2-receptor antagonist in the management of gastro esophageal reflux (Stanciu and Bennett, 1974), and as a haemostatic agent in surgical dressings (Thomas, 1990; Qin and Gilding, 1996). Alginate dressings, used to treat exuding wounds, often contain significant amounts of sodium alginate as this improves the gelling properties (Thomas, 2000). Sponges composed of sodium alginate and chitosan produce a sustained drug release and may be useful as wound dressings or as tissue engineering matrices (Lai et al., 2003). Sodium alginate is also used in cosmetics and food products (Table 2.5).

Table 2.5 Uses of sodium alginate

Use	Concentration (%)
Pastes and creams	5–10
Stabilizer in emulsions	1–3
Suspending agent	1–5
Tablet binder	1–3
Tablet disintegrant	2.5–10

Safety

Sodium alginate is widely used in cosmetics, food products, and pharmaceutical formulations, such as tablets and topical products, including wound dressings. It is generally regarded as a nontoxic and nonirritant material, although excessive oral consumption may be harmful. A study in five healthy male volunteers fed a daily intake of 175 mg/kg bodyweight of sodium alginate for 7 days, followed by a daily intake of 200 mg/kg body-weight of sodium alginate for a further 16 days, showed no significant adverse effects (Anderson et al., 1991).

Inhalation of alginate dust may be irritant and has been associated with industrial-related asthma in workers involved in alginate production. However, it appears that the cases of asthma were linked to exposure to seaweed dust rather than pure alginate dust (Henderson, et al., 1984).

LD_{50} (cat, IP): 0.25 g/kg (Lewis, 2004)

LD_{50} (mouse, IV): 0.2 g/kg

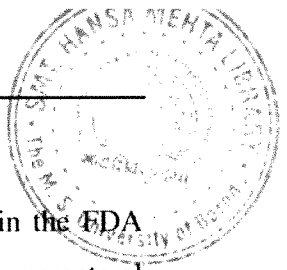
LD_{50} (rabbit, IV): 0.1 g/kg

LD_{50} (rat, IV): 1 g/kg

LD_{50} (rat, oral): >5 g/kg

Handling Precautions

Observe normal precautions appropriate to the circumstances and quantity of material handled. Sodium alginate may be irritant to the eyes or respiratory system if inhaled as dust. Eye protection, gloves, and a dust respirator are recommended. Sodium alginate should be handled in a well-ventilated environment.



Regulatory Status

GRAS listed. Accepted in Europe for use as a food additive. Included in the FDA Inactive Ingredients Guide (oral suspensions and tablets). Included in non parenteral medicines licensed in the UK. Included in the Canadian List of Acceptable Non-medicinal Ingredients.

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