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The application of mucoadhesive polymers in nasal drug delivery

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Abstract

Background: Over the last decades, the application of mucoadhesive polymers in nasal drug delivery systems has gained interest among pharmaceutical scientists as a means of promoting dosage form residence time in the nasal cavity as well as improving intimacy of contact with absorptive membranes of the biological system. In addition, the enhanced paracellular absorption following the swelling of the mucoadhesive polymers on the nasal membranes provides an important way for the absorption of the macromolecules through the nasal cavity. *Method*: This paper describes some aspects of mucoadhesion related to the nasal drug delivery system. First the theories of the adhesion of mucoadhesive polymers to the mucosa epithelium are described. Then the characteristics and application of several widely used polymers in nasal drug delivery are presented. Finally, the influences of dosage form on the nasal absorption of drugs in the mucoadhesive polymer-based formulations are discussed. *Conclusion*: The mucoadhesive polymers have enormous potential for the delivery of therapeutic macromolecules, genes, and vaccines through the nasal cavity with high drug bioavailability.

Key words: *Absorption enhancer; microspheres; mucoadhesion; mucociliary clearance; nanoparticles; nasal drug delivery; polymer*

Introduction

Conventionally the nasal cavity is used for the treatment of local diseases, such as rhinitis and nasal congestion. However, in the past decades nasal drug delivery has been paid much more attention as a promising drug administration route for the systemic therapy[1]. This is due to the anatomy and physiology of the nasal passage, such as the large surface area, highly vascularized epithelium, porous endothelial membrane, and the avoidance of first-pass metabolism[2].

Because of its ready accessibility, nasal drug administration has been considered as an alternative route for systemic use of drugs restricted to intravenous administration[3]. This is particularly important for the delivery of peptides and proteins that currently are mainly administered through intravenous route because of their susceptibility to the gastrointestinal proteases[4]. Nasal drug delivery can also provide a route of entry to the brain that circumvents the blood–brain barrier because the olfactory receptor cells are in direct contact with the central nervous system[5–9].

Currently nasally administered drugs focus on not only the treatment of acute diseases such as pain, panic attacks, sleep induction, erectile dysfunction, nausea, heart attacks, Parkinson's disease, but also the treatment of long-term illnesses such as diabetes, growth deficiency, osteoporosis, endometriosis, and hypertensions[10]. Recently the nasal mucosa is considered as an attractive site for the delivery of vaccines, not only because it has a relatively large absorptive surface and low proteolytic activity, but also because the nasal vaccines will improve the patients' compliance and reduce the production costs compared with the parenteral products. Increasing studies report that, when administered intranasally, vaccines can induce both local and systemic immune response[11,12].

Despite the high permeability of nasal membrane, generally only small molecular drugs (<1000 Da) show adequate absorption in the nasal cavity[13], most hydrophilic

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and macromolecular drugs such as insulin show low bioavailability or even no absorption at all[14]. The main reason for this is that they are lowly permeable and susceptible to the proteases in the nasal mucosal membrane, so these drugs can be rapidly cleared from the cavity, by ciliary movement or enzymatic degradation before they reach the bloodstream, and cannot cross the mucosal barriers[15]. Penetration enhancers such as surfactants[16], bile salts[17,18], fusidate derivatives[19], and phospholipids[20] have been used to improve the drug absorption through nasal mucosa, but toxicity tests proved that they were limited for clinical use because of their irreversible damage to nasal mucosa accompanied with their absorption-enhancing effects[21].

Some mucoadhesive polymers, such as cellulose, polyacrylate, starch, and chitosan, have proven to be effective on improving intranasal absorption of hydrophilic macromolecules. These polymers achieve this by increasing the drug residence time in the nasal cavity or enhancing intranasal absorption; some of them can serve both functions. Most of these polymers are generally recognized as safe (GRAS) pharmaceutical excipients and not absorbed, so they would not be expected to display systemic toxicity.

Even though a number of challenges are still to be overcome, the encouraging results stimulate pharmaceutical researchers to further efforts in order to develop new nasal formulations to replace the conventional parenteral products. In this article, the use of mucoadhesive polymers for the intranasal delivery of drugs is reviewed. Their ability of enhancing the intranasal absorption of macromolecular hydrophilic drugs will be focused on.

Mucoadhesion/bioadhesion

Definition

In 1986, Longer et al. defined the term 'bioadhesion' as 'the attachment of a synthetic or biological macromolecule to mucus and/or an epithelial surface for an extended period time'[22]. Similarly, Gu et al. described the term 'mucoadhesion' as 'the binding of polymers to mucin/epithelial surface'[23]. In nasal drug delivery, mucoadhesion means the adherence of a polymeric material to nasal epithelial surface (bioadhesion) or nasal mucus (mucoadhesion).

Mechanism of mucoadhesion

The process of mucoadhesion following nasal administration relates to the interaction between the mucoadhesive polymer and the mucus secreted by the submucosal glands[24]. The sequential events that occur during the mucoadhesion include the proper wetting and swelling of the polymer, and intimate contact between the polymer and the nasal mucosa. Then, the swelled mucoadhesive polymer penetrates into the tissue crevices followed by the interpenetration between the polymer chains and the protein chains of the mucus (Figure 2)[25].

To obtain sufficient absorption of drugs, firstly, the formulation should spread well on the nasal mucosa. Therefore, the spreadability is very important for the liquid mucoadhesive formulation, so do the flowability and wettablility for the solid mucoadhesive formulation[26,27].

Hydration of the polymer (swelling) plays a very important role in mucoadhesion, through which the polymer chains are liberated and interact with the biological tissue[28]. During hydration, there is a dissociation of hydrogen bonds of the polymer chains. When the polymer–water interaction becomes greater than the polymer–polymer interaction, adequate free polymer chains will be available for interaction between the polymer and the biological tissue[25]. The van der Waals, hydrogen, hydrophobic, and electrostatic forces between the polymer and the biological tissue (including the mucus), which form secondary chemical bonds, result in the adhesion of polymer to the mucosa[29,30]. There is a critical degree of hydration required for optimum mucoadhesion. The incomplete hydration because of the lack of the water leads to incomplete liberation of the polymer chains. On the other hand, an excessive amount of water will weaken the mucoadhesive bonds by overdiluting the polymer solution[31].

The polymer chains penetrating into the tissue crevices can hold back the ciliary movement, which will increase the retention time of the drugs in the nasal cavity[32]. Furthermore, the existence of the mucoadhesive carrier also reduces the contact between the drugs and the enzymes existing in the mucosa[25]. These both can enhance the intranasal absorption of hydrophilic drugs (the comparison of ordinary intranasal formulation with mucoadhesive intranasal formulation is showed in Figure 1). Apart from these, the dehydration of the epithelial cells after hydration may also temporarily open the tight junctions between the epithelial cells and improve the paracellular absorption of macromolecular drugs[31,32]. The opening mechanism has been demonstrated by the decrease in ZO-1 proteins and the change in the cytoskeletal protein F-actin from a filamentous to a globular structure[33]. This function of the mucoadhesive polymer is very important for the enhancement of the intranasal absorption of macromolecules weighing above 1000 Da[34].

Mucoadhesion can slow down the mucociliary clearance, but with time, mucus production will lead to the inordinate swelling of the mucoadhesive polymer and the reduction of the mucoadhesion bond strength, allowing a recovery of normal mucociliary movement rate and the clearance of the polymer from the mucosa[25].

Although many references indicate that the mucoadhesive polymer is effective in enhancing the intranasal absorption of macromolecular drugs, very few papers focus on the changes of gel structure and rheology of

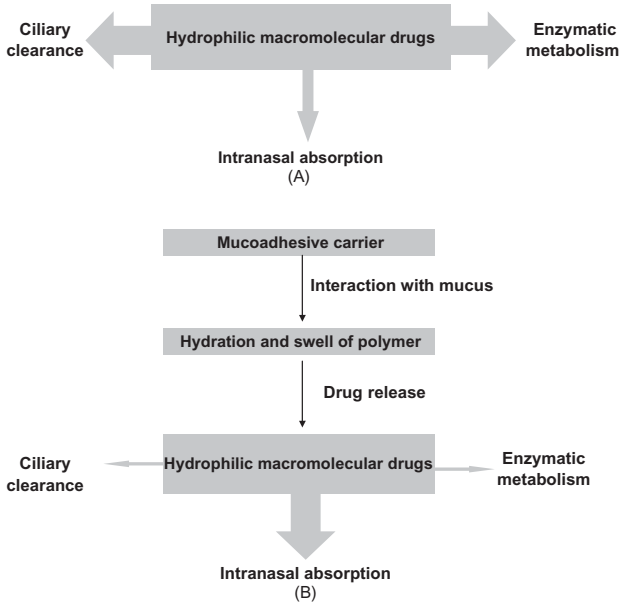

Figure 1. Schematic representation of the comparison of ordinary intranasal formulation with mucoadhesive intranasal formulation. (A) Ordinary intranasal delivery: little fraction of drugs can be absorbed because of the low permeability of the hydrophilic macromolecular drugs; most of drugs will be cleared by the ciliary movement or be metabolized by the enzymes existing in the nasal cavity. (B) Mucoadhesive intranasal drug delivery: the mucoadhesive carrier enhances the intranasal absorption by increasing the retention time of drugs and promoting the paracellular absorption in the nasal cavity whereas reducing the ciliary clearance. The mucoadhesive carrier can also protect the drugs from the enzymatic metabolism to a large extent.

the mucus caused by the mucoadhesive polymer and to what extent the interaction between the polymer and the mucus influences the release of the drugs, including in the disease condition. Disease conditions can affect mucoadhesion because of their influence on either mucus production or ciliary movement, and then may result in undesired drug release. Thus a good understanding of the nature of mucus in these diseases is imperative in designing a good nasal drug delivery system. Mucoadhesive capabilities of polymers should be studied under such disease conditions during the product development.

Mucoadhesive polymers used in nasal drug delivery

Cellulose derivatives

Cellulose is a class of most available polysaccharide, containing of 8000–10,000 glucose residues linked by β-1,4 glucosidic bonds[35]. There are many pharmaceutical grade derivatives of cellulose widely used in different administration routes. Several cellulose derivatives have proved to be effective in enhancing the intranasal absorption of drugs, including soluble cellulose derivatives such as hydroxypropyl methylcellulose (HPMC), hydroxypropyl cellulose (HPC), methylcellulose (MC) and carboxymethylcellulose (CMC), and insoluble cellulose derivatives such as ethylcellulose (EC) and microcrystalline cellulose (MCC). Table 1 summarizes the nasal drug delivery studies where the cellulose derivatives were employed as mucoadhesive carrier.

Cellulose derivatives can markedly prolong the residence time of drugs in the nasal cavity because of their desirable mucoadhesive property[43]. Additionally, because of their high viscosity following hydration in the nasal cavity, the celluloses can sustain the release of drugs[46]. For these reasons using celluloses as absorption enhancer can lead to improved intranasal absorption and increased bioavailability. Many references show that the celluloses are effective in increasing the intranasal bioavailability of small hydrophobic and hydrophilic macromolecular drugs (Table 1). For example, administered nasally with CMC, apomorphine can obtain a relative bioavailability of 102% compared with subcutaneous injection in rabbits[35]. Another study reported that an absolute bioavailability up to 90.77% could be achieved for ketorolac tromethamine administered with MCC[37]. The peptide drugs leuprolide and FD-4, when dosed with MCC/HPC through nasal route, reached an absolute bioavailability of 34.9% and 35.5% in rabbits, respectively[41].

Sometimes, combination of the celluloses with other absorption enhancer would obtain better effectiveness than using the polymer alone. Ozsoy et al. reported that the intranasal absolute bioavailability of ciprofloxacin in rabbits using MC and hydroxyethyl cellulose (HEC) alone as enhancer is only 18.2% and 19.46%, respectively. When combining with the Tween 80, the bioavailability increased to 22.35% and 25.39%, respectively[45]. In another study by Ikeda et al. on the intranasal delivery of dopamine, the combination of the HPC and azone led to an absolute bioavailability of almost 100% whereas it was only 25% for using HPC alone[42].

Polyacrylates

Polyacrylates have been investigated very frequently in many drug administration routes, such as oral[47], ocular[48], transdermal[49,50], and nasal[51] drug delivery systems, because of their excellent mucoadhesive and gel-forming capability. Among the pharmaceutical polyacrylates, carbomers and polycarbophil, which differ in the cross-linking condition and viscosity, are widely used in the nasal mucoadhesive drug delivery systems[52]. Table 2 summarized the studies on the use of polyacrylates in nasal drug delivery system.

Polyacrylates, capable of attaching to mucosal surfaces, can offer the prospects of prolonging the residence time of drugs at the sites of drug absorption and ensure intimate

Table 1. Summary of some nasal drug delivery studies where cellulose derivatives were employed.

Mucoadhesive polymer	Drugs	Dosage forms	Bioavailability (%)	Animal species	References
EC	FD-4	Powder	38.0 ± 3.8 (abs)	Rats	Ishikawa et al.[36]
CMC	Apomorphine	Powder	102 ± 15.6 (rel versus SC)	Rabbits	Ugwoke et al.[35]
MCC, pH 5.95	Ketorolac trom ethamine	Spray	90.77 (abs)	Rabbits	Quadir et al.[37]
MCC, pH 3.2	Ketorolac acid	Spray	63.89 (abs)	Rabbits	Quadir et al.[37]
MCC, pH 6.0	Ketorolac acid	Spray	70.6 (abs)	Rabbits	Quadir et al.[37]
MCC	Ketorolac acid	Powder	45 (abs)	Rabbits	Quadir et al.[37]
MCC	Ketorolac trom ethamine	Powder	38 (abs)	Rabbits	Quadir et al.[37]
MCC	Insulin	Spray	1.9 (abs)	Rabbits	Dondeti et al.[38]
MCC/sodium taurocholate	Insulin	Spray	8.36 (abs)	Rabbits	Dondeti et al.[38]
MCC/ammonium glycyrrhizinate	Insulin	Spray	7.83 (abs)	Rabbits	Dondeti et al.[38]
MCC	Cyanocobalamin	Powder	25.0 (abs)	Rabbits	Garcia-Arieta et al.[39]
MCC	Glucagon	Powder	—	Human	Teshima et al.[40]
MCC/HPC	Leuprolide	Powder	34.9 (abs)	Rabbits	Suzuki and Makino[41]
MCC/HPC	Calcitonin	Powder	16.4 (abs)	Rabbits	Suzuki and Makino[41]
MCC/HPC	FD-4	Powder	35.5 (abs)	Rabbits	Suzuki and Makino[41]
HPC	Dopamine	Liquid	25.0 (abs)	Dogs	Ikeda et al.[42]
HPC/azone	Dopamine	Liquid	100 (abs)	Dogs	Ikeda et al.[42]
HPC/Carbopol 934P/Poloxamer 407	Metoclopramide hydrochloride	Gel	51.0 (abs)	Rabbits	Zakia et al.[43]
HPMC/ sulfobutylether-β-cyclodextrin	Midazolam	Spray	73 (abs)	Human	Loftsson et al.[44]
HPMC	Ciprofloxacin	Gel	40.21 ± 6.41 (abs)	Rabbits	Ozsoy et al.[45]
MC	Ciprofloxacin	Gel	18.2 ± 4.8 (abs)	Rabbits	Ozsoy et al.[45]
MC/Tween 80	Ciprofloxacin	Gel	22.3 ± 5.5 (abs)	Rabbits	Ozsoy et al.[45]
HEC	Ciprofloxacin	Gel	19.46 ± 2.7 (abs)	Rabbits	Ozsoy et al.[45]
HEC/Tween 80	Ciprofloxacin	Gel	25.39 ± 2.19 (abs)	Rabbits	Ozsoy et al.[45]

abs, absolute; EC, ethyl cellulose; CMC, carboxymethylcellulose; FD-4, fluorescein isothiocyanate-labeled dextrans with an average MW of 4400; HEC, hydroxyethyl cellulose; HPC, hydroxypropyl cellulose; HPMC, hydroxypropylmethyl cellulose; MC, methyl cellulose; MCC, microclrystalline cellulose; rel, relative; SC, subcutaneous injection.

contact between the formulation and the membrane surface. Studies by Ugwoke et al. in rabbits reported that the use of Carbopol 971P in nasal dosage forms increases their residence time in the nasal cavity. The percentage of the formulations cleared from the nasal cavity at 3 hours was 24% for Carbopol 971P, whereas it was 70% for lactose[58]. Sustained release of drugs can also be obtained by using polyacrylates in nasal formulation, which result in a more stable blood concentration–time curve. Another research by Ugwoke et al. showed that the T_{max} of the Carbopol 971P-containing formulation of apomorphine was 52.21 minutes, which represented a fivefold improvement compared with that of the lactose-containing formulation, whereas the C_{max} of the Carbopol 971P-containing formulation was 330.2 ng/mL, lower than that of the lactose-containing formulation, which was 450.7 ng/mL[52].

Besides the mucoadhesion capability, polyacrylates may also temporarily open the tight junctions between the epithelial cells during the swelling progress in the nasal cavity and improve the paracellular absorption of drugs[55]. Based on these, polyacrylates can increase the intranasal bioavailability of both small hydrophobic drugs and hydrophilic macromolecular drugs. Using the Carbopol 971P and polycarbophil in the nasal apomorphine formulation, a relative drug bioavailability of 99.8% and 105.0% compared with subcutaneous injection could be obtained, respectively[53]. An absolute bioavailability of 14.4% in rabbits was reported for intranasal insulin formulation containing Carbopol 974P[56].

The Carbopol and polycarbophil are considered as GRAS by FDA, and many studies show that they are nonirritant to the skin and eye and nontoxic orally[52]. Callens et al. reported that the effect of Carbopol on the mucosa is negligible and reversible, no change of the epithelium barrier was observed even after a 4-week administration of Carbopol-based powder formulation in rabbits[56,59].

Table 2. Summary of some nasal drug delivery studies where polyacrylates were employed.

Mucoadhesive polymer	Drugs	Dosage forms	Bioavailability (%)	Animal species	References
Carbopol 971P	Apomorphine	Powder	99.8 ± 9.7 (rel versus SC)	Rabbits	Ugwoke et al.[53]
Polycarbophil	Apomorphine	Powder	105.0 ± 8.6 (rel versus SC)	Rabbits	Ugwoke et al.[53]
Carbopol 971P (15%, 30%, 60%)	Apomorphine	Powder	27,752.3 36,862.5 34,869.2[a]	Rabbits	Ugwoke et al.[52]
Carbopol 974P (15%, 30%)	Apomorphine	Powder	26,993.0 35,881.8[b]	Rabbits	Ugwoke et al.[52]
Polyarbophil	Apomorphine	Powder	28,831.4 32,894.9 34,415.9[c]	Rabbits	Ugwoke et al.[52]
Carbopol 934P	Fluorescein isothiocyanate	Powder	33 (abs)	Rabbits	El-Shafy et al.[54]
Carbopol 934P	Levonorgestrel	Liquid	99.4 (rel versus oral)	Rats	Shahiwala and Misra[55]
Carbopol 981P	Metoclopramide	Solution	17.48 (abs)	Sheeps	Tas et al.[51]
Carbopol 981P	Metoclopramide	Gel	56.79 (abs)	Sheeps	Tas et al.[51]
Carbopol 981P	Metoclopramide	Powder	23.29 (abs)	Sheeps	Tas et al.[51]
Carbopol 981P/DM-β-CD	Metoclopramide	Powder	31.16 (abs)	Sheeps	Tas et al.[51]
Carbopol 934P/HPC /Poloxamer 407	Metoclopramide hydrochloride	Gel	51.0 (abs)	Rabbits	Zaki et al.[43]
Carbopol 974P/DDWM	Insulin	Powder	14.4 ± 3.5 (abs)	Rabbits	Callens and Remon[56]
Carbopol 974P/Maltodextrin DE 8	Insulin	Powder	7.1 ± 1.6 (abs)	Rabbits	Callens and Remon[56]
Gelatin/polyacrylic microspheres	Oxyprenolol	Powder	–	Rats	Preda and Leucuta[57]

abs, absolute; DDWM, drum-dried waxy maize starch; DM-β-CD, dimethyl-β-cyclodextrin; HPC, hydroxypropyl cellulose; rel, relative; SC, subcutaneous injection. [a]Area under curve (ng mL/min) for formulations containing 15%, 30%, 60% Carbopol 971P, respectively. [b]Area under curve (ng mL/min) for formulations containing 15%, 30% Carbopol 974P, respectively. [c]Area under curve (ng mL/min) for formulations containing 15%, 30%, 60% polycarbophil, respectively.

Starch

The starch is one of the most widely used mucoadhesive carrier for nasal drug delivery, which has been reported to be effective on improving the absorption of both small hydrophobic drugs and hydrophilic macromolecular drugs (see Table 3). Maize starch is the most preferred class for pharmaceutical purpose, among which the drum-dried waxy maize starch (DDWM), because of its better bioadhesive property, has been considered as the best one compared with starch processed through other methods[56].

Starch can be used as nasal drug carriers in the form of powders, microspheres, or nanoparticles (Table 3), among which the degradable starch microspheres (DSM), also known as Spherex®, is the most widely used and also the first example of mucoadhesive microparticulate nasal delivery system[73]. These microspheres are prepared by an emulsion polymerization technique, in which the starch is cross-linked with epichlorohydrine and can incorporate molecules weighing less than 30 kDa[73].

Because of its mucoadhesion, the DSM can enhance the drug absorption by prolonging the residence time of drugs in the nasal cavity[74]. Illum et al. has observed that the half-life of clearance for DSM was prolonged to 240 minutes compared with 15 minutes for the liquid and powder control formulations[63]. Bjork and Edman suggested that water uptake by DSM and subsequent swelling might cause dehydration of the epithelial cells leading to the widening of tight junctions and as a consequence facilitate the paracellular transport of large hydrophilic molecules such as insulin[66]. It was suggested that the extent of drug absorption was improved even further when DSM were combined with the biological enhancers such as lysophosphatidylcholine (LPC)[60–62]. DSM can also protect the proteins wrapped in it against degradation by proteases in the mucosa. Several studies showed that the release of drugs from DSM was rapid and not sustained. This suggested that the utility of DSM in nasal drug delivery could further be exploited in the treatment of crisis diseases[35]. It was reported that DSM were well tolerated both in experimental animals and in humans; a test on healthy volunteers showed that only a small hyperplasia in the septum wall was observed when the DSM were administered two times per day for 8 weeks in dosages of 20 mg[64,75,76].

Chitosan

Chitosan [2-amino-2-deoxy-(1→4)-β-d-glucopyranan] is a linear cationic polysaccharide that is

Table 3. Summary of some nasal drug delivery studies where starch was employed.

Mucoadhesive polymer	Drugs	Dosage forms	Bioavailability (%)	Animal species	References
DSM	Apomorphine	Powder	96 ± 7.8 (rel versus SC)	Rabbits	Ugwoke et al.[35]
DSM	Desmopressin	Powder	4.7 ± 0.5 (abs)	Sheeps	Critchley et al.[60]
DSM/LPC	Desmopressin	Powder	9.6 ± 2.8 (abs)	Sheeps	Critchley et al.[60]
DSM	Gentamicin	Powder	50 (abs)	Rats	Illum et al.[61]
DSM	Gentamicin	Powder	9.7 (abs)	Sheeps	Illum et al.[61]
DSM/LPC	Gentamicin	Powder	57.3 (abs)	Sheeps	Illum et al.[61]
DSM	HGH	Powder	2.7 (rel versus SC)	Sheeps	Illum et al.[62]
DSM/LPC	HGH	Powder	14.4 (rel versus SC)	Sheeps	Illum et al.[62]
DSM	Insulin	Liquid	240/15[a]	Human	Illum et al.[63]
DSM	Insulin	Powder	30–40[b]	Human	Edman et al.[64]
DSM	Insulin	Powder	4.5 (abs)/10.7 (rel versus SC)	Sheeps	Farraj et al.[20]
DSM/LPC	Insulin	Powder	13.1 (abs)/31.5 (rel versus SC)	Sheeps	Farraj et al.[20]
DSM	Insulin	Powder	30 (rel versus SC)	Rats	Björk and Edman[65]
DSM	Insulin	Powder	10 (abs)	Rats	Björk and Edman[66]
DSM	Insulin	Powder	3.6 (rel versus SC)	Sheeps	Illum et al.[18]
DSM/GDC/STDHF	Insulin	Powder	31.9 (rel versus SC)	Sheeps	Illum et al.[18]
DSM/STDHF	Insulin	Powder	16.5 (rel versus SC)	Sheeps	Illum et al.[18]
DSM	Melatonin	Powder	84.07 (abs)	Rabbits	Mao et al.[67]
DDWM	Insulin	Powder	4.46 (abs)	Rabbits	Callens and Remon[56]
DDWM/Carbopol 974P	Insulin	Powder	14.4 ± 3.5 (abs)	Rabbits	Callens and Remon[56]
Amioca starch/Carbopol 974P	Insulin	Powder	17.8 ± 4.5 (abs)	Rabbits	Callens et al.[68]
DDSM/Carbopol 974P	Insulin	Powder	13.4 ± 3.2 (abs)	Rabbits	Callens et al.[68]
DSM	Metoclopramide	Liquid	137 (rel versus SC)	Human	Vivien et al.[69]
POG-nanoparticles/Na glyco	Insulin	Powder	23.4 ± 2.7 (rel versus SC)	Rats	Jain et al.[70]
POE-nanoparticles/Na glyco	Insulin	Powder	21.7 ± 3.5 (rel versus SC)	Rats	Jain et al.[70]
EG-nanoparticles/Na glyco	Insulin	Powder	22.6 ± 4.1 (rel versus SC)	Rats	Jain et al.[70]
EE-nanoparticles/Na glyco	Insulin	Powder	35.9 ± 5.8 (rel versus SC)	Rats	Jain et al.[70]
POG-nanoparticles/LPC	Insulin	Powder	20.6 ± 3.3 (rel versus SC)	Rats	Jain et al.[70]
POE-nanoparticles/LPC	Insulin	Powder	17.3 ± 2.7 (rel versus SC)	Rats	Jain et al.[70]
EG-nanoparticles/LPC	Insulin	Powder	21.2 ± 3.5 (rel versus SC)	Rats	Jain et al.[70]
EE-nanoparticles/LPC	Insulin	Powder	25.2 ± 4.2 (rel versus SC)	Rats	Jain et al.[70]
EE-L1-nanoparticles/Na glyco	Insulin	Powder	26.1 ± 5.0 (rel versus SC)	Rats	Jain et al.[70]
EE-L2-nanoparticles/Na glyco	Insulin	Powder	44.3 ± 7.6 (rel versus SC)	Rats	Jain et al.[70]
EE-L3-nanoparticles/Na glyco	Insulin	Powder	35.9 ± 5.8 (rel versus SC)	Rats	Jain et al.[70]
SMS	G-CSF	Powder	1.7 ± 1.1 (rel versus SC)	Sheeps	Jabbal-Gill et al.[109]
SMS/LPC	G-CSF	Powder	8.4 ± 3.4 (rel versus SC)	Sheeps	Jabbal-Gill et al.[109]
SMS	Morphine hydrochloride	Powder	74.8 ± 29.2 (abs)	Sheeps	Illum et al.[71]
Starch	Insulin	Powder	19.2 ± 5.3 (abs)	Rabbits	Pringle et al.[72]

abs, absolute; DDWM, drum-dried waxy maize starch; DSM, degradable starch microspheres; EE-nanoparticles, starch nanoparticles prepared by emulsion method using epichlorohydrin as cross-linker, L1, L2, L3, epichlorohydrin volume used is 0.1, 0.2, 0.3 mL, respectively; EG-nanoparticles, starch nanoparticles prepared by gel method using epichlorohydrin as cross-linker; HGH, human growth hormone; GDC, deoxyglycocholate; LPC, L-a-lysophosphatidylcholine; Na glyco, sodium glycocholate; POE-nanoparticles, starch nanoparticles prepared by emulsion method using $POCl_3$ as cross-linker; POG-nanoparticles, starch nanoparticles prepared by gel method using $POCl_3$ as cross-linker; rel, relative; SC, subcutaneous injection; SMS, starch microsphere; STDHF, sodium taurodihydrofusidate. [a]Half-life clearance (minutes) of starch microspheres/solution formulation in the nasal cavity. [b]The time (minutes) for occurrence of maximal reduction of plasma glucose.

obtained by a process of deacetylation from chitin, an abundant structural polysaccharide in shells of crustacea such as lobsters, shrimps, and crabs[77]. Because of the NH_2 groups resultant from the deacetylation process chitosan is insoluble at neutral and alkaline pH. However, it can form water-soluble salts with inorganic and organic acids including glutamic acid, hydrochloric acid, lactic acid, and acetic acid. Toxicity tests have revealed that the LD_{50} of chitosan in mice exceeds 16 g/kg[78]. Because of its low cost, biodegradability, and biocompatibility, chitosan has been increasingly applied as pharmaceutical excipients in oral, ocular, nasal, implant, parenteral, and transdermal drug delivery[79].

Chitosan and its derivatives have been shown to be active in enhancing the intranasal drug absorption because of their excellent mucoadhesive properties. It was also confirmed that coating micro- and nanoparticulates with chitosan could improve drug adsorption to mucosal surfaces[80]. Besides their hydration in the nasal cavity, the interaction of the positively charged amino group with the negatively charged sites on the mucosa surface also contributes to their mucoadhesion[77]. Soane et al.[81] reported that chitosan microspheres and solutions resulted in three- and eightfold longer clearance half-lives compared with sodium pertechnetate solution in sheep nasal cavity, respectively. In addition, many studies have proved that chitosan and its derivatives could transiently open the tight junctions between the cells and lead to the paracellular transport of drugs[82,83]. Table 4 summarized the recent nasal drug delivery studies where chitosan derivatives were employed as absorption enhancer.

Chemical and biological properties of chitosan, such as mucoadhesion and ability in enhancing nasal absorption, are determined by the types of derivatives, degree of deacetylation, and molecular weight (MW). Because chitosan is only soluble in acidic environment in which the amino groups at the C-2 position are protonated. At neutral pH, most chitosan molecules will lose their charge and precipitate from solution. Recent studies have shown that only protonated, soluble chitosan can trigger the opening of tight junctions and thereby facilitate the paracellular transport of hydrophilic mannitol[101]. To improve the poor water solubility of chitosan, some derivatives were synthesized, such as trimethyl chitosan[102,103] and polyethylene glycol (PEG)-chitosan[86]. Thanou et al. reported that the trimethyl chitosan was soluble and effective in enhancing intranasal absorption even at neutral pH[102]. It was reported that 5-methylpyrrolidinone chitosan[104], thiolated chitosan[85], and *N*-trimethyl chitosan hydrochloride[99] are more mucoadhesive than unmodified chitosans and show a higher bioavailability in vivo compared with the unmodified chitosans.

Mei et al. reported that the permeation-enhancing effect of chitosan increased with increasing MW up to 100 kDa[84]. Study by Tengamnuay et al. suggested that chitosans should differ in their MW by at least twofold in order to have a clearly differentiating effect on the nasal absorption enhancement of a kyotorphin analogue[105]. On the contrary, Zaki et al. found that there is no significant difference between the constants of intranasal absorption for metoclopramide HCl administered with chitosan high weight (600 kDa) and low weight (150 kDa) even though they differ in MW by fourfold[100]. The same result was obtained in study by Aspden et al.[106].

Because of the positive charge of chitosan in a weak acidic environment, it can also be applied to deliver the negatively charged DNA through nasal mucosa and protect them from nuclease degradation[107]. Compared with viral vectors, this alternative vector markedly reduced the safety risks resulting in high transfectability[108]. Recently many studies showed that nasal immunization with chitosan plus inactive vaccine is a potentially effective, easily administered form of vaccination. *Bordetella pertussis* filamentous hemagglutinin and recombinant pertussis toxin have shown to induce very strong systemic and mucosal immune reactions against the antigens when intranasally administrated with chitosan[109, 110].

Read et al. confirmed that the standard inactivated trivalent influenza vaccine administered intranasally in combination with chitosan glutamate (0.5%, w/w) could induce both systemic and local immune responses, and the results were not statistically different from those obtained following administration of the commercial influenza vaccine by the intramuscular route[111]. Bacon et al.[112] have reported that chitosan solutions are able to enhance both the mucosal and the systemic immune responses against influenza virus vaccines. Only in mice that received chitosan/vaccine formulation intranasally, high IgA titers in nasal washings could be found. This was not observed in mice receiving the antigen through subcutaneous injection[112].

Other cationic macromolecular materials, such as poly-L-arginine and aminated gelatin have also been investigated for their application as nasal absorption enhancers[98,99]. These polymers work in a similar way to chitosan, at least in animal models, and have been found to be effective in enhancing the absorption of fluorescein isothiocyanate (FITC)-dextran and insulin with only negligible nasal toxicity[97–99] (also been listed in Table 4).

Dosage forms

Because of their good physical characteristics the mucoadhesive polymers can be used as absorption enhancer for nasal drug delivery in liquid spray, solid powder, or particulate formulation. However, the dosage form to a large extent influences the efficiency of the polymer enhancer. The polymers can be rendered ineffective or mediocre by the choice of an inappropriate dosage form. On the other hand, by selecting an optimal dosage form the polymers can be more effective on improving the pharmacokinetics of the drugs. Different dosage forms based on the same polymer may result in absorption of drugs of different extent, by affecting the physiological conditions of the nasal cavity (mucociliary clearance, membrane structure, enzymatic activity) and the drug behavior in the nasal cavity (deposition site, residence time, drug release rate, transmembrane modes). Therefore, developing an appropriate polymer-based formulation suitable for the treatment of diseases is very important.

Table 4. Summary of some nasal drug delivery studies where chitosan derivatives and other positively charged macromolecules were employed.

Mucoadhesive polymer	Drugs	Dosage forms	Bioavailability	Animal species	References
Chitosan (50 kDa)	2,3,5,6-tetramethylpyrazine	Liquid	57.3 ± 8.7 (abs)	Rats	Mei et al.[84]
Chitosan (100 kDa)	2,3,5,6-tetramethylpyrazine	Liquid	68.4 ± 3.1 (abs)	Rats	Mei et al.[84]
Trimethylated chitosan (50 kDa)	2,3,5,6-tetramethylpyrazine	Liquid	73.6 ± 6.5 (abs)	Rats	Mei et al.[84]
Thiolated chitosan (100 kDa)	2,3,5,6-tetramethylpyrazine	Liquid	65.1 ± 5.2 (abs)	Rats	Mei et al.[84]
Chitosan	Insulin	Liquid	9–15 (rel versus SC)	Human	Illum[79]
Thiolated chitosan microspheres	Insulin	Powder	7.24 ± 0.76 (abs)	Rats	Krauland et al.[85]
Chitosan microspheres	Insulin	Powder	2.04 ± 1.33 (abs)	Rats	Krauland et al.[85]
Chitosan-PEG nanoparticles	Insulin	Liquid	46%/17%[a]	Rabbits	Zhang et al.[86]
Chitosan glutamate	Human growth hormone	Powder	14 ± 9 (rel versus SC)	Sheeps	Cheng et al.[87]
Chitosan microspheres	Insulin	Powder	44 (abs)	Rats	Varshosaz et al.[88]
Chitosan	Levonorgestrel	Liquid	101.7 (rel versus oral)	Rats	Shahiwala and Misra[55]
Chitosan	Salmon calcitonin	Liquid	201.2 (rel versus IN plain drug)	Rats	Sinswat and Tengamnuay[89]
Chitosan glutamate	Insulin	Liquid	3.6 ± 0.8 (rel versus SC)	Sheeps	Dyer et al.[90]
Chitosan glutamate	Insulin	Powder	17.0 ± 6.6 (rel versus SC)	Sheeps	Dyer et al.[90]
Chitosan complex	Insulin	Liquid	1.8 ± 0.9 (rel versus SC)	Sheeps	Dyer et al.[90]
Chitosan nanoparticles	Insulin	Liquid	1.3 ± 0.8 (rel versus SC)	Sheeps	Dyer et al.[90]
Chitosan (1.5%)	Insulin	Liquid	15.4 ± 5.4 (rel versus SC)	Rats	Yu et al.[91]
Chitosan (1.0%)	Insulin	Liquid	11.3 ± 5.3 (rel versus SC)	Rats	Yu et al.[91]
Chitosan/EDTA	Insulin	Liquid	8.8 ± 4.5 (rel versus SC)	Rats	Yu et al.[91]
Chitosan/Tween 80	Insulin	Liquid	11.8 ± 3.9 (rel versus SC)	Rats	Yu et al.[91]
Chitosan glutamate	Carbamazepine	Powder	800/25[b]	Sheeps	Gavini et al.[92]
Chitosan microsphere	Goserelin	Liquid	40 (abs)	Sheeps	Lim et al.[93]
Chitosan microspheres	Pentazocine	Powder	96.5 ± 8.4 (abs)	Rabbits	Sankar et al.[94]
Chitosan glutamate	Morphine hydrochloride	Solution	26.6 ± 14.5 (abs)	Sheeps	Illum et al.[71]
Chitosan microspheres	Morphine hydrochloride	Powder	54.6 ± 28.8 (abs)	Sheeps	Illum et al.[71]
Chitosan glutamate	Morphine hydrochloride	Liquid	56.0 ± 27.0 (abs)	Human	Illum et al.[71]
Chitosan glutamate	Morphine hydrochloride	Powder	56.0 ± 20.0 (abs)	Human	Illum et al.[71]
Chitosan	Gentamicin	Powder	31.4 ± 2.7 (abs)	Rabbits	Lim et al.[95]
Chitosan/hyaluronan	Gentamicin	Powder	42.9 ± 3.5 (abs)	Rabbits	Lim et al.[96]
Chitosan free amine	Calcitonin	Liquid	2.45 (abs)	Rats	Sinswat and Tengamnuay [89]
Gelatin microspheres	Levodopa	Powder	—	Rats	Brime et al.[97]
Chitosan	FITC-TP 5	Liquid	46.05 ± 0.11 (abs)	Rats	Wang et al.[98]
Aminate gelatin microspheres	Insulin	Liquid	5.0 (abs)	Rats	Wang et al.[98]
Aminate gelatin microspheres	Insulin	Powder	8.6 ± 2.9 (abs)	Rats	Wang et al.[98]
Poly-L-arginine 10 (5.0%)	FD-4	Liquid	54.5 (abs)	Rats	Miyamoto et al.[99]
Poly-L-arginine 50 (2.0%)	FD-4	Liquid	94.7 (abs)	Rats	Miyamoto et al.[99]
Poly-L-arginine 100 (2.0%)	FD-4	Liquid	97.9 (abs)	Rats	Miyamoto et al.[99]
Chitosan	—	Solution/ microspheres	43/115/15[c]	Sheeps	Soane et al.[81]
Chitosan	Metoclopramide	Liquid	0.06983/0.07517/0.0452[d]	Rats	Zaki et al.[17]
Chitosan	Metoclopramide	Spray	87.2 ± 7.7 (abs)	Rabbits	Zaki et al.[100]

FD-4, fluorescein isothiocyanate-labeled dextrans with an average MW of 4400; FITC-TP 5, fluorescein isothiocyanate (FITC)-labeled thymopentin (TP 5). [a]Reduction of the plasma glucose after administration of insulin-PEG-chitosan nanoparticle/insulin PBS solution. [b]C_{max} (ng/mL) ratio of microspheres/pure drug. [c]Half-time of clearance of chitosan solution/chitosan microsphere/sodium pertechnetate solution (control). [d]Constant of absorption (min^{-1}) of the formulation containing chitosan low weight/high weight and control plain drug solution.

Solution

The solution dosage form is easy to prepare and convenient to use. Nowadays, as more metered dose nasal actuators are available, the nasal solutions are most commonly delivered through metered dose nasal actuator systems. These systems can precisely deliver actuation volumes as low as 25 μL[19]. The solution formulations based on the mucoadhesive polymers have been shown to increase the drug bioavailability by increasing the residence time of the drug at the nasal mucosa[44,91,99].

However, only the low-dose drug or highly soluble drug is administrable in a simple solution dosage form because of the small volume of the nasal cavity[36]. Moreover the viscosity of solution for intranasal administration is limited to approximately 500 mPas, preparation with higher viscosity allows better mucoadhesion, but its instillation to the nasal cavity is more difficult[113]. The stability issue may also be a problem for solution, in many cases; especially for the peptide and protein formulations, preservatives should be added in the solution formulation, which may cause morphological changes in adenoid tissue in chronic use[114].

Dried powder

Polymer-based powder formulations show no adhesion until their absorption of mucus occurs on the nasal mucosa surface. This allows easy application to the nasal cavity by metered dose insufflation even if the polymer is highly mucoadhesive. In addition, liquid preparations are more easily cleared to the nasopharynx and oropharynx from where they enter the posterior part of the tongue[10]. Therefore administration of nasal powders may increase patient compliance, especially if the smell and taste of the delivered drug is unacceptable.

After getting in contact with the nasal mucosa, polymer-based powders are believed to form a viscous gel following absorbing water from the nasal mucus (Figure 2). Then the free polymer chains penetrating into the tissue crevices can hold back the ciliary movement, which will increase the retention time of the drugs in the nasal cavity[25].

Compared with solution, powder formulation can facilitate administering larger amounts of drugs and result in a higher drug concentration on the mucosa surface, which leads to the saturation of the enzymatic degradation and then raise the drug bioavailability[114]. Dry powder formulations can also avoid the utilization of preservatives and freezed storage, because they do not support microbial growth and are more stable than solution[115]. For these reasons, the dried powder is the most commonly studied formulation for the nasal drug delivery, including small hydrophobic drugs, peptide drugs, and vaccine. Garmise et al.[116] prepared dry powder nasal influenza vaccine formulation by using spray-freeze-drying method; the results indicated the powders were amorphous and more stable with respect to liquid formulations. In vivo experiments demonstrated that the powders significantly increased residence time in rats and elicited enhanced serum and mucosal antibody response[116].

Methods of preparing powders include physical mixture, co-ground, co-spray, and co-lyophilization. Polymer-based powders prepared using different

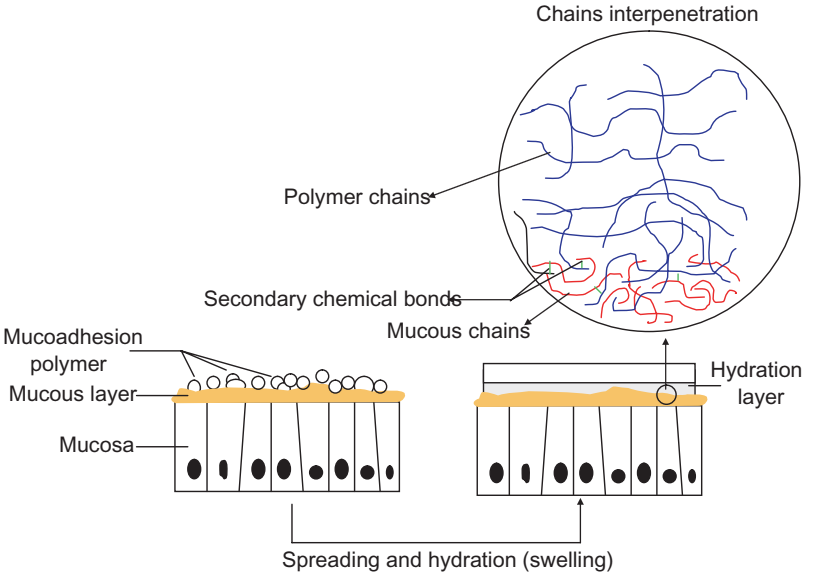

Figure 2. Schematic representation of the process of mucoadhesion on the nasal mucosa surface.

methods might display different properties, such as the density, flow ability, crystal type, drug release characteristics, and capability in enhancing the nasal absorption of drugs[1]. Ascentiis et al. found that the progesterone-β-cyclodextrin powder prepared by co-ground showed a significantly slower drug release rate and more prolonged drug level than the same powder manufactured by co-lyophilization[117]. Another research showed that physical powder mixture prepared by mixing insulin powder with DDWM and Carbopol 974P displayed significantly lower bioavailability than the free-dried powders[56].

Although polymer-based powder formulations display many advantages in nasal administration, several problems should be paid attention to in designing of nasal powder formulations. Powders may cause irritation on the nasal mucosa and give a gritty feel to the tissue[19]. In addition, it is known that intranasal deposition of particles depends on their aerodynamic size; size control of intranasally administered powders will guarantee their deposition in the respiratory region, where the maximum of absorption occurs. Particles smaller than 10 μm in diameter risk pulmonary deposition whereas those exceeding 50 μm might be deposited in the anterior part of the nose, which is covered by non-ciliated squamous epithelium and is unfavorable for absorption[118]. It will be difficult and cost consuming to manufacture powders with specified particle size.

Gel

It is well known that gels can promote an intimate contact between formulations and the mucosa surface and prolong the residence time by reducing the ciliary clearance rate[119]. Studies by Tas et al. showed that the nasal bioavailability of metoclopramide in gel formulation prepared by using Carbopol 981P was higher than that in solution and lyophilized powder formulation based on the same polymer[51]. However, because of its high viscosity the gel is difficult to administer and an accurate drug dose cannot be measured.

To overcome this problem the nasal in situ gel has been developed, i.e., thermosensitive or pH-sensitive gel[43,120]. The in situ gel is fluid-like before nasal administration, which is convenient for administration and accuracy of drug dosing. After the formulation contact with the mucosa, the special temperature or pH value of the mucus promotes the transition from liquid to gel, which prolongs drug residence times and improves drug bioavailability.

Zaki et al. prepared an in situ gel of metoclopramide hydrochloride with solution-gel transition temperature of about 25–32°C by using poloxamer 407 as thermogelling moderator[43]. Results of rat experiment showed that this in situ gel prolonged the mucociliary transport time from 10 to 52 minutes (compared with sodium chloride) and maintained nasal mucosal integrity after 14 days of application[43]. The bioavailability study in rabbits revealed that the absolute bioavailability of metoclopramide hydrochloride was significantly increased from 51.7% in case of the oral drug solution to 69.1% in case of the nasal in situ gel[43].

Ghosh et al. designed intranasal in situ gel systems of sumatriptan with a gelation temperature below 34°C using thermoreversible polymer Pluronic F127 and mucoadhesive polymer Carbopol 934P[120]. The results of in vitro drug permeation studies across sheep nasal mucosa indicate that the in situ gelling formulation was effective in improving the permeation coefficient, and the histopathological examination did not detect any damage during in vitro permeation studies[120].

Wu et al. prepared thermosensitive hydrogel of insulin by simply mixing *N*-[(2-hydroxyl-3-trimethylammonium) propyl] chitosan chloride and PEG with a small amount of a-b-glycerophosphate. The solution-gel transition temperature of this hydrogel is about 34°C[121]. In vivo experiment demonstrated that the hydrogel formulation decreased the blood glucose concentration by 40–50% for at least 4–5 hours after administration, and no apparent cytotoxicity was found after application[121].

Nasal particulate systems

Nasal particulate systems using mucoadhesive polymers as carriers include microparticle/sphere and nanoparticle. Particulate drug carrier systems administered through nasal mucosa may protect the drug from enzymatic degradation, increase the drug dissolution rate, intensify the contact of the formulation with the mucosa, enhance the uptake by the epithelium, and act as a controlled release system resulting in prolonged blood concentrations[122,–124]. Among the polymers widely used as nasal drug particulate carrier, the positively charged polymers such as chitosan and aminated gelatin are most attractive because of their hydrogel nature, which leads to opening of the tight junctions and their intimate contact with the negatively charged mucosa membrane[98,99]. In vivo evaluation in rabbits has proved that chitosan nanoparticles were able to improve the nasal absorption to a great extent compared with chitosan solution because of the intensified contact of the nanoparticle with the nasal mucosa as compared with chitosan solutions[122].

It has been believed that nanoparticles possess superiority over microspheres as nasal drug carrier because their larger surface area results in more intimate contact with the mucosa, which leads to higher local concentration gradient[70]. Moreover, nanoparticles cross the mucosal epithelium better than microspheres do. Microparticles smaller than 10 μm

administered intranasally are believed to be taken up by the M-cells overlaying the nasal associated lymphoid tissue and transported to sub-mucosal layers[124]. However, in case of the nanoparticles, besides the M-cells and associated phagocytosis, the epithelial cells are also involved in the transport of nanoparticles by internalization[125].

Recent study by Amidi et al. showed that FITC-albumin-loaded chitosan nanoparticles, when administered in the nasal cavity, were able to cross the mucosal layer, taken up by rat nasal epithelia and nasal associated lymphoid tissue cells. This property of nanoparticles provides a good indication of their potential as gene and vaccine carriers[126]. Teijeiro-Osorio et al. found that the transfection efficiency of the nanoparticles loaded with pSEAP (100–200 nm) was higher than the naked DNA (control)[127].

Recently, many studies confirmed that association of vaccines to the nanoparticulate systems has shown to enhance the antigen uptake by nasal lymphoid tissues[128]. By incorporating the vaccine into nanoparticulates, the vaccine is protected against degradation on its way to the mucosal tissue and efficiently taken up by the M-cells; subsequently the carriers are biodegraded and the antigen releases in the Peyer's patches and inducing strong systematic and mucosal immune responses against the antigens[129,130]. For this reason, nasal particulate vaccine systems are preferred to conventional intramuscular products for the prophylaxis of some upper respiratory tract infection such as influenza, pertussis, and diphtheria[1].

Conclusion

With advantages such as mucoadhesion, an increase in the residence time of the polymer, penetration enhancement, and enzymatic inhibition, mucoadhesive polymers will undoubtedly be utilized for the nasal delivery of a wide variety of therapeutic compounds. This class of polymers has enormous potential for the delivery of therapeutic macromolecules, genes, and vaccines. Unfortunately, only a few studies have been conducted with new-generation mucoadhesive polymers for nasal drug delivery, and very few papers focus on the changes of structure and rheology of the mucus caused by the mucoadhesive polymer, to what extent the interaction between the polymer and the mucus influences the release of the drugs including in the disease condition. With recent advancements in the fields of biotechnology and cytoadhesion, the authors believe that there will be both academic and industrial efforts to explore this new area of nasal drug delivery, and it might not be too far fetched to envisage more and more nasal products that employ mucoadhesive polymers.

Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of this paper.

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