Influence of Intranasal Drugs on Human Nasal Mucociliary Clearance and Ciliary Beat Frequency

Jian Jiao 1,2, Luo Zhang 1,2*

1Department of Otolaryngology, Head and Neck Surgery, Beijing Tongren Hospital, Capital Medical University, Beijing, China
2Beijing Key Laboratory of Nasal Diseases, Beijing Institute of Otolaryngology, Beijing, China

ABSTRACT

The nasal mucociliary clearance system, which comprises epithelial cilia and mucus from goblet cells, is an important intrinsic defense mechanism of the upper respiratory tract. Intranasal drugs and additives can have a detrimental effect on ciliary activity and mucociliary clearance, and thus impact the integrity of nasal defense mechanisms. This article discusses the current literature on the effects of different classes of intranasal drugs including intranasal corticosteroids, antihistamines, decongestants, antimicrobials and antivirals, as well as various drug excipients and nasal irrigation solutions on human nasal mucociliary clearance and ciliary beat frequency. Available data indicate that some intranasal formulations tend to hamper nasal ciliary function and mucociliary clearance. Therefore, it is of great importance to assess the effects of intranasal drugs and additives on mucociliary function before they are recommended as therapy for different nasal conditions.

Keywords: Intranasal administration; excipients; nasal irrigations; mucociliary clearances; cilia

INTRODUCTION

Intranasal administration of drugs has generated much attention within pharmaceutical industry as a viable option for local or systemic delivery of diverse therapeutic compounds in recent years. The intranasal route provides several advantages for drug delivery due to the noninvasive nature of the process, a large nasal mucosa surface area, rapid onset of therapeutic effect, potential for direct-to-central nervous system delivery, avoidance of first-pass metabolism, and likelihood for maximal patient comfort and compliance. 1 A prerequisite for intranasal formulations, however, is that the drugs and additives should not interfere with normal nasal function, especially mucociliary clearance function.

The nasal mucociliary clearance system, which comprises epithelial cilia and mucus from goblet cells, is one of the most important nonspecific defense mechanisms of the respiratory tract. Under normal conditions, epithelial cilia beat in a coordinated and unidirectional fashion to transport mucus through the epithelium to various drainage sites, and thus remove inhaled particles and irritants such as dust, bacteria, viruses and air pollutants from the airway. 2 Inhibition of the mucociliary clearance system induces a longer contact time of nasal mucus with entrapped particles and irritants, possibly leading to airway infection and
Mathematical modeling shows that energy transferred by cilia to mucus blanket is proportional to the square of ciliary beat frequency (CBF). Additionally, experimental data have demonstrated that a relatively modest increase in CBF (16%) resulted in a 56% increase in surface liquid velocity, i.e., mucociliary transport. Thus, CBF is an important parameter for assessing ciliary function and mucociliary clearance.

Evidence-based international guidelines have recommended the use of intranasal corticosteroids, antihistamines, decongestants, anti-infective agents and nasal irrigation for the treatment of rhinitis, sinusitis, and related allergic or chronic nasal conditions. Intranasal drug formulations are composed of active drugs and various formulation excipients such as preservatives and absorption enhancers, which may individually or in combination have harmful effects on nasal ciliary function or mucociliary clearance. This article reviews the current literature on the effects of different types of intranasal formulations on human nasal mucociliary clearance and CBF.

### EFFECTS OF INTRANASAL CORTICOSTEROIDS

Intranasal corticosteroids are recommended as first-line therapy for the treatment of allergic rhinitis due to their efficacy, tolerability and ease of use. However, debates over the safety of intranasal corticosteroids and their potential side effects on nasal mucociliary function have been on-going for several years. Some of the current, widely used intranasal corticosteroids include budesonide, fluticasone propionate/furoate, triamcinolone acetonide and mometasone furoate as well as newer non-aqueous intranasal corticosteroid aerosols beclomethasone dipropionate and ciclesonide.

Several studies have investigated the influence of intranasal corticosteroids on mucociliary clearance. An early study by Holmberg and Pipkorn reported that topical beclomethasone dipropionate suspension treatment did not change mucociliary clearance in healthy volunteers, as indicated by utilizing the saccharine-dye test. Similarly, Klossek and colleagues and Pata and colleagues have employed the saccharine test to assess the effect of 6 months’ treatment with triamcinolone acetonide or 1 month’s treatment with mometasone furoate, respectively, and demonstrated that neither compound impaired mucociliary function in patients with perennial allergic rhinitis. Employing a radiotracer technique, Naclerio and colleagues compared the influence of 2 weeks’ treatment with either budesonide or mometasone on nasal mucociliary function and reported that neither drug impaired mucociliary clearance function (Table 1).

However, the effects of intranasal corticosteroids on ciliary motility observed in vitro are different from those observed in vivo. Early studies have indicated that while budesonide appeared to decrease human nasal CBF only slightly in vivo; beclomethasone dipropionate and flunisolide induced a dose-related, irreversible decrease in human nasal CBF in vitro. Using cultured human nasal epithelial cells, Hofmann and colleagues reported that budesonide spray did not affect CBF at 10% dilution and induced moderate reversible decrease in CBF at 50% dilution. In contrast, fluticasone propionate and mometasone furoate sprays induced a reversible decrease in CBF at 10% dilution and a complete, irreversible ciliostasis at 50% dilution. Using primary human nasal epithelial culture models, we have previously demonstrated that budesonide induced a rapid but reversible ciliostasis at undiluted therapeutic concentrations and a gradual but fully reversible decrease in CBF.
at 50% dilution, whereas no effect was observed on CBF at 10% dilution. In contrast, fluticasone propionate induced irreversible ciliostasis when used undiluted and at up to 50% dilution of therapeutic concentration, whereas there was a reversible decrease of CBF at 10% dilution (Table 1).

### EFFECTS OF INTRanasAL ANTIHISTAMINES

Intranasal antihistamines, along with intranasal corticosteroids, are also proposed as first-line therapy in patients with allergic rhinitis. In contrast to intranasal corticosteroids, intranasal antihistamines have a more rapid onset of action, ranging from 15 to 30 minutes, and are therefore especially useful in patients with episodic nasal symptoms or as a pretreatment before inhaled allergen exposure. Antihistamines are functionally classified as first- or second-generation antihistamines according to whether or not they enter blood brain barrier readily and cause side effects such as sedation and mucosal dryness. Current widely used second-generation antihistamines, for example azelastine and levocabastine, cause fewer adverse side effects and are also highly potent and selective H1-receptor antagonists, with many of these compounds possessing additional antiallergic and anti-inflammatory properties.

Studies investigating the influence of intranasal antihistamines on mucociliary clearance and ciliary function have provided contradictory results (Table 2). Some studies have demonstrated no adverse effects of intranasal formulations containing azelastine or levocabastine on mucociliary clearance and/or ciliary function, either in vivo or in vitro. In contrast, others have reported that intranasal formulations containing azelastine and levocabastine do produce ciliotoxic effects on human nasal epithelium in vivo. We have also assessed the effects of azelastine and levocabastine nasal sprays on CBF of human nasal epithelial cell cultures and demonstrated that while undiluted aqueous azelastine or levocabastine caused irreversible ciliostasis and a 10% dilution resulted in reversible decreases in CBF of these cultures, up to 5% dilution of these formulations had no effect on the CBF of these cultures.

| Compound            | Indicator     | Effect                        |
|---------------------|---------------|-------------------------------|
| Azelastine          | CBF           | No effect/Decrease, irreversible |
| Levocabastine       | CBF           | No effect/Decrease, reversible |
|                     | Mucociliary clearance | No effect |

CBF, ciliary beat frequency.

### Table 1. Effects of intranasal corticosteroids on human nasal mucociliary clearance and CBF

| Compound                          | Indicator         | Effect                          |
|-----------------------------------|-------------------|---------------------------------|
| Beclomethasone dipropionate       | Mucociliary clearance | No effect/Decrease, irreversible |
| Triamcinolone acetonide           | Mucociliary clearance | No effect/Decrease, irreversible |
| Mometasone furoate                | Mucociliary clearance | No effect/Decrease, irreversible |
| Budesonide                        | Mucociliary clearance | No effect/Decrease, reversible |
| Fluticasone propionate            | CBF               | Decrease, irreversible          |

CBF, ciliary beat frequency.

### Table 2. Effects of intranasal antihistamines on human nasal mucociliary clearance and CBF

| Compound          | Indicator | Effect                        |
|-------------------|-----------|-------------------------------|
| Azelastine        | CBF       | No effect/Decrease, irreversible |
| Levocabastine     | CBF       | No effect/Decrease, irreversible |
|                   | Mucociliary clearance | No effect |

CBF, ciliary beat frequency.

---

https://e-aair.org

https://doi.org/10.4168/aair.2019.11.3.306
EFFECTS OF INTRANASAL DECONGESTANTS

Intranasal decongestants are the most powerful drugs available for the reduction of nasal obstruction. These drugs cause vasoconstriction and reduce congestion of the mucosa in favour of the available volume of nasal cavities for air passage and conditioning, and are therefore widely used as an adjuvant for improving ventilation of sinuses in the treatment of allergic rhinitis and acute or chronic rhinitis. Intranasal decongestants can be divided into 2 groups: sympathomimetic amines (e.g., adrenaline, phenylephrine, ephedrine and phenylpropanolamine) and imidazoline derivatives (e.g., naphazoline, oxymetazoline and xylometazoline). Serving as commonly used nasal decongestants; ephedrine is a nonselective \( \alpha/\beta \)-adrenergic receptor agonist, while phenylephrine, naphazoline, oxymetazoline and xylometazoline are selective \( \alpha \)-adrenergic receptor agonists.

The influence of intranasal decongestants on human nasal mucociliary clearance and CBF is summarized in Table 3. Teng and colleagues\(^24\) demonstrated that ephedrine (0.5% and 1%) decreased the human mean nasal mucociliary transport rate in healthy volunteers.\(^24\) In contrast to these findings, studies from our laboratory investigating the effects of ephedrine on the regulation of human nasal CBF have demonstrated ephedrine to induce instant and moderate increases in CBF followed by inhibitory responses.\(^25\) Moreover, we demonstrated that the clinically used concentration of ephedrine (0.5%) induced the highest stimulatory effect, without any obvious inhibitory effect on human nasal CBF.\(^25\) Phillips and colleagues\(^26\) investigated the \textit{in vivo} and \textit{in vitro} effect of phenylephrine on nasal CBF and mucociliary transport of healthy volunteers and demonstrated that while phenylephrine had a cilio-stimulatory effect \textit{in vivo} as well as at a low concentration (0.01%) \textit{in vitro}, it also had a cilio-inhibitory effect at higher concentrations (0.25% and 0.5%) \textit{in vitro}.\(^26\) Similarly, Min and colleagues\(^27\) reported that at concentrations of 0.125% to 2.5%, phenylephrine induced a significant decrease in human nasal CBF in a dose-dependent- and time-dependent manner.\(^27\)

Oxymetazoline, xylometazoline and naphazoline have also been reported to inhibit ciliary movement and mucociliary transport in human nasal mucosa.\(^28\)–\(^30\) Mickenhagen and colleagues\(^31\) have investigated the influence of different alpha-sympathomimetic drugs on the CBF of cultured human nasal mucosa cells \textit{in vitro}. They demonstrated that while oxymetazoline was not ciliotoxic at concentrations of 0.01% and 0.001%, it did cause irreversible damage to cilia and a significant decrease in CBF, which was partially reversible, at a concentration of 0.1%. In contrast, naphazoline at concentrations ranging from 0.001% to 0.1% was not shown to significantly alter CBF.\(^31\) We have also investigated the effects of oxymetazoline on CBF and mucociliary transport time in cultured nasal mucosal tissue of

| Compound       | Indicator          | Effect Description                                                                 |
|----------------|--------------------|------------------------------------------------------------------------------------|
| Ephedrine      | Mucociliary clearance | Decrease\(^24\) Instant increase followed by decrease\(^30\) |
| Phenylephrine  | Mucociliary clearance | Increase\(^16\) Increase (low concentration), decrease (high concentration)\(^29\) / Decrease\(^22\) |
| Oxymetazoline  | Mucociliary clearance | Decrease, reversible\(^25\)/Increase\(^29\)/No effect (low concentration), decrease (high concentration)\(^34\) Decrease\(^29\)/No effect (low concentration), decrease (high concentration)\(^22\) |
| Xylometazoline | CBF                | Decrease\(^29\)/Decrease, partially reversible\(^29\) |
| Naphazoline    | CBF                | Decrease\(^29\)/No effect\(^31\) |

CBF, ciliary beat frequency.
healthy individuals and demonstrated that CBF was not significantly altered at concentrations of oxymetazoline 0.025% or 0.05%, but was significantly decreased at concentrations of oxymetazoline 0.10% and 0.20%. Additionally, 0.05% oxymetazoline increased the mean human nasal mucociliary transport time. Another study has investigated the effects of nasal decongestants on the mucociliary transport rate and ciliary structure in the human nasal mucosa of healthy volunteers using the saccharine test and electron microscopy, respectively and reported that while 0.025% or 0.05% oxymetazoline neither affected mucociliary transport nor the ciliary structure in the nasal mucosa, higher concentrations of 0.5% and 1.0% ephedrine significantly decreased mucociliary transport as well as exfoliated the cilia in the nasal mucosa of these individuals.

**EFFECTS OF INTRANASAL ANTIMICROBIALS AND ANTIVIRALS**

Topical anti-infective therapy is a promising addition to chronic rhinosinusitis treatment, particularly for patients with persistent or recurrent disease. Current anti-infective therapy includes antibacterials, antifungals, antivirals, etc. Mallants and colleagues investigated the effects of different antibiotics on CBF of human nasal epithelial cells and demonstrated that clarithromycin and neomycin did not influence ciliary activity, while bacitracin, clindamycin, gramicidin and roxithromycin significantly increased CBF. However, another investigation of the effects of some protease inhibitors on human nasal CBF reported that bacitracin (8 mM), as well as puromycin (135 mM) had no effect on CBF after acute exposure, but significantly reduced the CBF following 15-minute daily exposure for 1 week. Indeed, several other antibacterials, including ofloxacin, betadine and mupirocin, have also been reported to decrease ciliary activity in human nasal epithelial cells. A recent study investigating the influence of nebulized drugs on nasal ciliary activity reported that nebulization of tobramycin, colistimethate, ceftazidime and aztreonam did not affect CBF, while a tobramycin-containing solution manufactured for intravenous use had a negative effect on CBF.

With respect to the influence of antivirals on ciliary function and mucociliary clearance, Han and colleagues have reported that although 20 mg/mL ribavirin, a broad spectrum antiviral agent, did not influence ciliary activity in vitro; 50 mg/mL ribavirin slowed ciliary beating significantly and 60 mg/mL caused ciliostasis. Nasal inhalation of ribavirin at 60 mg/mL for up to 20 minutes, however, neither slowed mucociliary clearance, nor affect the ciliary beat of nasal epithelium examined in vitro immediately after inhalation. Another study investigating the effect of ribavirin on ciliary activity, however, showed that a concentration of 500 mg/mL ribavirin did not affect ciliary beating of nasal epithelial cells collected from either individuals with symptomatic colds or healthy volunteers. Dimova and colleagues investigated the effects of 5,7,3′,4′-tetrahydroxy-3-O-methylflavone (3-MQ), an anti-rhinoviral compound for nasal application, on CBF of human nasal epithelial cells and demonstrated that 3-MQ at 2 mg/mL and 10 mg/mL had a reversible cilio-stimulatory effect, without any observable ciliotoxic effect at even higher concentrations up to 20 mg/mL.

Several antifungal drugs, including amphotericin B (AMB), clotrimazole and itraconazole, have also been reported to have an inhibitory effect on human nasal CBF. However, a study on the safety of a novel formulation of nanodisc (ND) containing super aggregated AMB (ND-AMB) for sinonasal delivery has recently demonstrated apically administered 75
μg/mL ND-AMB or AMB solution for 15 minutes to insignificantly alter the CBF of human nasal epithelial cells grown as air-liquid interface cultures. More recently, Jiang and colleagues evaluated the efficacy of AMB nasal irrigation as adjuvant therapy after functional endoscopic sinus surgery (ESS) on chronic rhinosinusitis patients, and demonstrated that nasal irrigation with 200 μg/mL AMB for 2 months did not significantly alter saccharine transit time, compared to pre-irrigation. The influence of different antimicrobials and antivirals on human nasal mucociliary clearance and CBF is summarized in Table 4.

### EFFECTS OF INTRANASAL DRUG EXCIPIENTS

Absorption enhancers and preservatives are commonly used drug excipients and indispensable components of intranasal drugs. Intranasal absorption enhancers are used to increase nasal membrane permeability so as to enhance efficient nasal absorption of the active drugs, while preservatives are essential for preventing microbial contamination, which could result as a consequence of repeated administration of the aqueous nasal formulation. Despite their effectiveness, some of these excipients have been reported to be toxic to the nasal epithelium and may interfere with mucociliary clearance and ciliary function. Thus, it is necessary to choose safe and effective excipients for intranasal drug delivery.

Many compounds of widely varying chemical structures, including bile acids, fusidate derivatives, fatty acids, phospholipids, cyclodextrins and chitosans, have been investigated as potential nasal absorption enhancers. Ideally absorption enhancers should not interfere with the normal mucociliary function of nasal mucosa; however, early studies demonstrated

| Compound               | Indicator | Effect                                           |
|------------------------|-----------|--------------------------------------------------|
| Clarithromycin         | CBF       | No effect                                        |
| Neomycin               | CBF       | No effect                                        |
| Bacitracin             | CBF       | Increase/No effect (acute exposure), decrease (1-week exposure) |
| Clindamycin            | CBF       | Increase                                        |
| Gramicidin             | CBF       | Increase                                        |
| Roxithromycin          | CBF       | Increase                                        |
| Puromycin              | CBF       | No effect (acute exposure), decrease (1-week exposure) |
| Ofloxacin              | CBF       | Decrease                                        |
| Betadine               | CBF       | Decrease                                        |
| Mupirocin              | CBF       | Decrease                                        |
| Tobramycin             | CBF       | No effect                                        |
| Colistimethate         | CBF       | No effect                                        |
| Ceftazidime            | CBF       | No effect                                        |
| Aztreonam              | CBF       | No effect                                        |
| Ribavirin              | CBF       | No effect (low concentration), decrease (high concentration)/No effect |
| 5,7,3′,4′-tetrahydroxy-3-O-methylflavone | CBF | Increase, reversible (low concentration), no effect (high concentration) |
| Amphotericin B         | CBF       | Decrease/No effect/No effect                     |
| Clotrimazole           | CBF       | Decrease                                        |
| Itraconazole           | CBF       | Decrease                                        |

CBF, ciliary beat frequency.

Intranasal Drugs and Nasal Mucociliary Clearance

https://e-aair.org

https://doi.org/10.4168/aair.2019.11.3.306

311
several absorption enhancers, such as polysorbate 80 and LPC, had a cilio-inhibitory effect in vitro. Various chitosans with different molecular weights were also reported to inhibit the mucociliary transport rate in human nasal tissue ex vivo, while once daily application of 0.25% chitosan solution for 7 days had no effect on saccharin clearance times in vivo. In contrast to these earlier compounds, cyclodextrins are now preferred absorption enhancers because of advantageous properties for improved drug solubilization, protection against physicochemical and enzymatic degradation, and the potential for improved absorption. Uchenna and colleagues assessed the effects of a series of cyclodextrins, including gamma-cyclodextrin, hydroxypropyl-beta-cyclodextrin, anionic-beta-cyclodextrin polymer, dimethyl-beta-cyclodextrin and alpha-cyclodextrin, on CBF using human nasal epithelial cell suspension cultures. They demonstrated that irreversible cilio-inhibition by cyclodextrins occurred only at concentrations exceeding those used in pharmaceutical formulations and/or after unusual exposure times in this model. Considering that dilution and mucociliary clearance contribute to a further decrease in local drug concentration in vivo, they proposed that cyclodextrins were safe nasal absorption enhancers (Table 5).

Thiolization of well-established polymers such as poly(acrylates) or chitosans by immobilization of sulfhydryl bearing ligands on the polymeric backbone results in production of “thiomers” with significantly improved mucoadhesive enzyme- and efflux-pump inhibiting, as well as permeation-enhancing properties. Palmberger and colleagues investigated the effects of gel formulations of thiomers on CBF in human nasal epithelial cells and found that Poly(acrylic acid) 450 kDa-cysteine (PAA-cys), alginate-cysteine (alg-cys) and chitosan-thiobutylamidine (chito-TBA) exhibited a concentration-dependent and partially reversible cilio-inhibitory effect. Thus, they suggested that thiomers were likely to be suitable excipients for nasal drug delivery systems, taking into account that dilution after application and cilio-modifying effects are usually more pronounced under in vitro conditions.

A variety of preservatives have been used in aqueous nasal formulations, such as benzalkonium chloride (BKC), potassium sorbate (PS), phenylethyl alcohol, chlorbutol, thiomersal, methylparaben and propylparaben. BKC is by far the most commonly used preservative in aqueous nasal formulations. However, the safety concerns about BKC remain controversial, particularly as some studies have shown that BKC caused impairment of CBF.

### Table 5. Effects of intranasal drug excipients on human nasal mucociliary clearance and CBF

| Compound                        | Indicator            | Effect                                      |
|---------------------------------|----------------------|---------------------------------------------|
| Polysorbate 80                  | CBF                  | Decrease, reversible<sup>41</sup>           |
| LPC                             | CBF                  | Decrease<sup>47</sup>                       |
| Chitosans                       | Mucociliary clearance| No effect<sup>48</sup>                      |
| Gamma-cyclodextrin              | CBF                  | No effect<sup>46</sup>                      |
| Hydroxypropyl-beta-cyclodextrin | CBF                  | No effect<sup>46</sup>                      |
| Alpha-cyclodextrin              | CBF                  | No effect (30-minute exposure), decrease (high concentration, after 45-minute exposure), partially reversible<sup>46</sup> |
| Dimethyl-beta-cyclodextrin      | CBF                  | No effect (30-minute exposure), decrease (high concentration, after 45-minute exposure), irreversible<sup>46</sup> |
| Poly(acrylic acid) 450 kDa-cysteine | CBF              | Decrease, partially reversible<sup>50</sup> |
| Alginate-cysteine               | CBF                  | Decrease, partially reversible<sup>50</sup> |
| Chitosan-thiobutylamidine       | CBF                  | Decrease, partially reversible<sup>50</sup> |
| Benzalkonium chloride           | CBF                  | Decrease, irreversible<sup>16,31,51</sup>   |
| Potassium sorbate               | Mucociliary clearance| Decrease<sup>31</sup>/No effect<sup>44</sup> |

CBF, ciliary beat frequency.
mucociliary clearance or degenerative changes in nasal mucosa,\textsuperscript{16,31,32} whereas others have reported BKC to have no toxic effects on the nasal mucosa.\textsuperscript{33,34} Nevertheless, it is important to note that while data generated \textit{in vitro} raises concerns regarding the safety of BKC, \textit{in vivo} data generally indicates BKC to be safe. Taking into account that the toxic effect of BKC \textit{in vitro} may be partially attenuated by absorption and dilution of respiratory mucus \textit{in vivo}, the use of BKC in intranasal formulations has therefore generally been considered to be safe. Similarly, PS is another commonly used preservative in nasal formulations. In contrast to BKC, studies on the effects of PS have not found the cilio-toxic effect of this compound at clinically used concentrations.\textsuperscript{35} Indeed, studies from our laboratory have also confirmed the safety of PS on CBF in human nasal ciliated cells.\textsuperscript{36} Thus, like BKC, PS can also be considered as a safe preservative for use in intranasal formulations (\textbf{Table 5}).

\section*{EFFECTS OF NASAL IRRIGATION}

Nasal irrigation is a simple, safe, effective therapeutic procedure that has been used in the treatment of nasal diseases for many years. Together with corticosteroid/pharmacological treatment, nasal irrigation is also recommended as first-line treatment in acute and chronic rhinosinusitis and after sinonasal surgery as well as adjunctive treatment for allergic rhinitis, acute upper respiratory tract infections, rhinitis in pregnancy, etc.\textsuperscript{35} Nasal irrigation may be effective in reducing nasal congestion and mucopurulent secretion, stimulating cleansing of the nasal and paranasal cavities, and in preventing crusting and moisturizing the mucosa after endonasal surgery. Nasal irrigation also appears to improve the mucociliary transport function of the nasal mucosa.\textsuperscript{36}

Different kinds of nasal irrigation solutions, such as normal saline as well as various concentrations of hypertonic saline, Ringer-Lactate solution, isotonic and hypertonic seawater solution have been used in clinical practice (\textbf{Table 6}). Saline solutions have been widely used in nasal irrigation for many years and are recommended for the treatment of various nasal diseases by several international expert groups.\textsuperscript{35,37} While the majority of studies on the effects of saline solution of different osmolarities on mucociliary clearance have reported hypertonic saline to be more effective than normal saline in improving mucociliary clearance,\textsuperscript{56,58,59} few studies have found no difference between hypertonic and normal saline.\textsuperscript{60,61} Interestingly, a study by Ural and colleagues\textsuperscript{62} has reported that irrigation with hypertonic saline restored impaired mucociliary clearance in chronic sinusitis patients, while isotonic saline improved mucociliary clearance in allergic rhinitis and acute sinusitis patients, suggesting that nasal irrigation with isotonic or hypertonic saline may improve mucociliary clearance time in various nasal pathologies.\textsuperscript{62} Studies on the effects of saline

| Compound               | Indicator            | Effect          |
|------------------------|----------------------|-----------------|
| Hypertonic saline      | Mucociliary clearance| Increase\textsuperscript{56,58,60,62}/No effect\textsuperscript{61},64 |
|                        | CBF                  | Decrease\textsuperscript{54,64} |
| Isotonic saline        | Mucociliary clearance| Increase\textsuperscript{56,60,62}/No effect\textsuperscript{61,64} |
|                        | CBF                  | Decrease\textsuperscript{69}/No effect\textsuperscript{44} |
| Ringer-Lactate solution| Mucociliary clearance| Increase\textsuperscript{62}/No effect\textsuperscript{64} |
| Hypertonic seawater    | Mucociliary clearance| Increase\textsuperscript{62} |
|                        | CBF                  | Decrease\textsuperscript{60} |
| Isotonic seawater      | CBF                  | Increase\textsuperscript{69} |
| Dexpanthenol in seawater| Mucociliary clearance| Increase\textsuperscript{62} |

CBF, ciliary beat frequency.
solutions of different osmolarities on CBF in vitro have indicated that while 0.9% normal saline did not affect or had a moderately negative effect on CBF of human nasal epithelium, an increase in saline tonicity was associated with increased inhibition of CBF, reversible ciliostasis or irreversible ciliostatic effect, depending on saline hypertonicity.\textsuperscript{63,64} Considering the findings from both in vivo and in vitro studies, isotonic or hypertonic saline solutions at concentrations of 2%–3% may be most appropriate for nasal irrigation.

Some studies have compared the effects of Ringer-Lactate solution with saline solution for nasal irrigation (Table 6). One study by Unal and colleagues\textsuperscript{65} found that patients who used Ringer-Lactate solution for irrigation after nasal septal surgery had a significantly better mucociliary transport time than patients using isotonic saline solution.\textsuperscript{65} In a more recent study by Low and colleagues\textsuperscript{66} conducted a double-blind, randomized controlled trial to investigate the effects of normal saline, lactated Ringer’s and hypertonic saline nasal douching solutions for 6-weeks after ESS. They reported that there was no significant improvement in mucociliary clearance with either of these solutions; however, irrigation with Ringer-Lactate solution resulted in significantly better improvement in sinonasal symptoms scores compared to normal or hypertonic saline solutions.\textsuperscript{66} They suggest that nasal irrigation with lactated Ringer’s solution after ESS may be better than irrigation with saline.

In recent years, isotonic or hypertonic seawater solution has commonly been used for nasal irrigation because of high levels of minerals and trace elements, such as calcium, potassium, magnesium and zinc ions, which can assist in epithelial wound repair and ciliary beat regulation. Indeed, Süslü and colleagues\textsuperscript{67} have reported that in patients who underwent septoplasty, 20 days’ nasal irrigation with hypertonic seawater significantly improved mucociliary clearance to a greater extent than isotonic saline irrigation, as indicated using the saccharine test.\textsuperscript{67} Similarly, Fooanant and colleagues\textsuperscript{68} have shown that dexpanthenol in seawater spray resulted in better mucociliary clearance than saline irrigation in sinusitis patients following ESS.\textsuperscript{68} More recently, Bonomet and colleagues\textsuperscript{69} conducted a randomized, controlled, blinded, in vitro study to assess the effect of normal saline and diluted or non-diluted seawater on CBF and epithelial wound repair speed (WRS) in airway epithelial cells from 13 nasal polyps explants. They demonstrated that non-diluted seawater enhanced the CBF and WRS of nasal epithelial cells when compared to normal saline and diluted seawater.\textsuperscript{69} Furthermore, undiluted seawater significantly enhanced CBF and slightly improved WRS compared to control medium. However, contrary to these findings, Laberko and colleagues\textsuperscript{70} have demonstrated that hypertonic seawater solution had a strong ciliotoxic effect on nasal ciliary epithelium in vitro\textsuperscript{70} (Table 6).

CONCLUSIONS

Nasal mucociliary clearance is an important intrinsic defense mechanism of the respiratory tract. A review of the published literature indicates that intranasal drugs including corticosteroids, antihistamines, decongestants, antimicrobials and antivirals, and various drug excipients such as preservatives and absorption enhancers as well as different nasal irrigation solutions may influence nasal mucociliary clearance or nasal mucosal ciliary function. Depending on whether intranasal drugs speed up, slow down or block mucociliary clearance or ciliary function, they subsequently influence different aspects of clinical activity. Thus, increased mucociliary clearance could lead to the drug being rapidly cleared and may result in much shorter duration and lower nasal bioavailability, whereas slowing down or
blockage of normal mucociliary clearance would result in longer contact of the nasal mucosa with entrapped viruses and bacteria, possibly leading to airway infection and damage to the mucosa. Therefore, it is of great importance to assess the effects of nasal drugs and additives on nasal ciliary function and mucociliary clearance in the development of new nasal drugs and the selection of appropriate safe excipients. However, it should be noted that some of the observed effects of drugs on CBF in vitro are different and more pronounced than their actual effects on ciliary activity and mucociliary clearance in vivo. Thus, if a compound does not affect mucociliary clearance in vivo or is shown to have no effect or only a mild effect in vitro, it can be considered safe for in vivo use.

REFERENCES

1. Costantino HR, Illum L, Brandt G, Johnson PH, Quay SC. Intranasal delivery: physicochemical and therapeutic aspects. Int J Pharm 2007;337:1-24. PUBMED | CROSSREF
2. Gudis DA, Cohen NA. Cilia dysfunction. Otolaryngol Clin North Am 2010;43:461-72. PUBMED | CROSSREF
3. Seybold ZV, Mariassy AT, Stroh D, Kim CS, Gazeroğlu H, Wanner A. Mucociliary interaction in vitro: effects of physiological and inflammatory stimuli. J Appl Physiol (1985) 1990;68:1421-6. PUBMED | CROSSREF
4. Brożek JL, Bousquet J, Baena-Cagnani CE, Bonini S, Canonica GW, Casale TB, et al. Allergic Rhinitis and its Impact on Asthma (ARIA) guidelines: 2010 revision. J Allergy Clin Immunol 2010;126:466-76. PUBMED | CROSSREF
5. Cheng L, Chen J, Fu Q, He S, Li H, Liu Z, et al. Chinese Society of Allergy guidelines for diagnosis and treatment of allergic rhinitis. Allergy Asthma Immunol Res 2018;10:300-53. PUBMED | CROSSREF
6. Benninger MS, Ahmad N, Marple BF. The safety of intranasal steroids. Otolaryngol Head Neck Surg 2003;129:739-50. PUBMED | CROSSREF
7. Raphael GD, Berger WE, Prenner BM, Finn AF Jr, Kelley L, Tantry SK. Efficacy, safety, and optimal dose selection of beclomethasone dipropionate nasal aerosol for seasonal allergic rhinitis in adolescents and adults. Curr Med Res Opin 2013;29:1329-40. PUBMED | CROSSREF
8. Mohar D, Berger WE, Laforce C, Raphael G, Desai SY, Huang H, et al. Efficacy and tolerability study of ciclesonide nasal aerosol in patients with perennial allergic rhinitis. Allergy Asthma Proc 2012;33:19-26. PUBMED | CROSSREF
9. Holmberg K, Pipkorn U. Influence of topical beclomethasone dipropionate suspension on human nasal mucociliary activity. Eur J Clin Pharmacol 1986;30:625-7. PUBMED | CROSSREF
10. Klossek JM, Laliberté F, Laliberté MF, Moutedji N, Bousquet J. Local safety of intranasal triamcinolone acetonide: clinical and histological aspects of nasal mucosa in the long-term treatment of perennial allergic rhinitis. Rhinology 2001;39:17-22. PUBMED
11. Pata YS, Akbaş Y, Unal M, Görür K, Ozcan C, Vayıoğlu Y. The effect of mometasone furoate on mucociliary clearance in patients with perennial allergic rhinitis. Kulak Burun Bogaz Ihtis Derg 2003;11:97-9. PUBMED
12. Naclerio RM, Baroody FM, Bidani N, De Tineo M, Penney BC. A comparison of nasal clearance after treatment of perennial allergic rhinitis with budesonide and mometasone. Otolaryngol Head Neck Surg 2003;128:220-7. PUBMED | CROSSREF
13. Duchateau GS, Zuiderma J, Merkus FW. The in vitro and in vivo effect of a new non-halogenated corticosteroid - budesonide - aerosol on human ciliary epithelial function. Allergy 1986;41:260-5. PUBMED | CROSSREF
14. Stafanger G. In vitro effect of beclomethasone dipropionate and flunisolide on the mobility of human nasal cilia. Allergy 1987;42:507-11. PUBMED | CROSSREF
15. Hofmann T, Gugatschga M, Koidl B, Wolf G. Influence of preservatives and topical steroids on ciliary beat frequency in vitro. Arch Otolaryngol Head Neck Surg 2004;130:440-5.

16. Jiao J, Meng N, Zhang L. The effect of topical corticosteroids, topical antihistamines, and preservatives on human ciliary beat frequency. ORL J Otorhinolaryngol Relat Spec 2014;76:127-36.

17. Horak F, Ziegelmayer UP, Ziegelmayer R, Kavina A, Marschall K, Munzel U, et al. Azelastine nasal spray and desloratadine tablets in pollen-induced seasonal allergic rhinitis: a pharmacodynamic study of onset of action and efficacy. Curr Med Res Opin 2006;22:151-7.

18. Zhang L, Cheng L, Hong J. The clinical use of cetirizine in the treatment of allergic rhinitis. Pharmacology 2013;92:14-25.

19. Achterrath-Tuckermann U, Saano V, Minker E, Stroman F, Arny I, Joki S, et al. Influence of azelastine and some selected drugs on mucociliary clearance. Lung 1992;170:201-9.

20. Merkus FW, Schüsler-van Hees MT. Influence of levocabastine suspension on ciliary beat frequency and mucociliary clearance. Allergy 1992;47:230-3.

21. Hofmann T, Wolf G, Koidl B. Effect of topical corticosteroids and topical antihistaminics on ciliary epithelium of human nasal mucosa in vitro. HNO 1998;46:146-51.

22. Albert J, Stoll W. The effect of antiallergic intranasal formulations on ciliary beat frequency of human nasal epithelium in vitro. Allergy 1998;53:986-9.

23. Passáli D, Salerni L, Passáli GC, Passáli FM, Bellussi L. Nasal decongestants in the treatment of chronic nasal obstruction: efficacy and safety of use. Expert Opin Drug Saf 2006;5:783-90.

24. Teng Y, Zhang X, Xu G, Cai Q, Xu J. The observation of the ciliotoxicity of nasal mucosa with nasal decongestant. Lin Chuang Er Bi Yan Hou Ke Za Zhi 2005;19:824-6.

25. Zhang L, Han D, Song X, Wang H, Wang K, Liu Z. Effects of ephedrine on human nasal ciliary beat frequency. ORL J Otorhinolaryngol Relat Spec 2008;70:91-6.

26. Phillips PP, McCaffrey TV, Kern EB. Third place--Resident Clinical Science Award 1990. The in vivo and in vitro effect of phenylephrine (Neo Synephrine) on nasal ciliary beat frequency and mucociliary transport. Otolaryngol Head Neck Surg 1990;103:558-65.

27. Min YG, Yun YS, Rhee CS, Sung MW, Lee KS, Ju MS, et al. Effects of phenylephrine on ciliary beat in human nasal respiratory epithelium: quantitative measurement by video-computerized analysis. Laryngoscope 1998;108:418-21.

28. Hofmann T, Wolf G, Koidl B. In vitro studies of the effect of vasoconstrictor nose drops on ciliary epithelium of human nasal mucosa. Laryngorhinootologie 1995;74:564-7.

29. Curtis LN, Carson JL. Computer-assisted video measurement of inhibition of ciliary beat frequency of human nasal epithelium in vitro by xylometazoline. J Pharmacol Toxicol Methods 1992;28:1-7.

30. Armengot M, Basterra J, Garcia-Bartual E. The influence of anesthetics and vasoconstrictors on nasal mucociliary transport. Acta Otorhinolaryngol Belg 1989;43:149-56.

31. Mickenhagen A, Siefer O, Neugebauer P, Stennert E. The influence of different alpha-sympathomimetic drugs and benzalkoniumchlorid on the ciliary beat frequency of in vitro cultured human nasal mucosa cells. Laryngorhinootologie 2008;87:30-8.

32. Zhang L, Han D, Song X, Wang K, Wang H. Effect of oxymetazoline on healthy human nasal ciliary beat frequency measured with high-speed digital microscopy and mucociliary transport time. Ann Otol Rhinol Laryngol 2008;117:127-33.
33. Mallants R, Jorissen M, Augustijns P. Beneficial effect of antibiotics on ciliary beat frequency of human nasal epithelial cells exposed to bacterial toxins. J Pharm Pharmacol 2008;60:437-43.

34. Remigius UA, Jorissen M, Willems T, Kinget R, Verbeke N. Mechanistic appraisal of the effects of some protease inhibitors on ciliary beat frequency in a sequential cell culture system of human nasal epithelium. Eur J Pharm Biopharm 2003;55:283-9.

35. Gosepath J, Grebneva N, Mossikhin S, Mann WJ. Topical antibiotic, antifungal, and antiseptic solutions decrease ciliary activity in nasal respiratory cells. Am J Rhinol 2002;16:25-31.

36. Birk R, Aderhold C, Wenzel A, Eschenhagen T, Kramer B, Hörmann K, et al. Mupirocin reduces ciliary beat frequency of human nasal epithelial cells. Eur Arch Otorhinolaryngol 2016;273:4335-41.

37. Kim JH, Rimmer J, Mrad N, Ahmadzada S, Harvey RJ. Betadine has a ciliotoxic effect on ciliated human respiratory cells. J Laryngol Otol 2015;129 Suppl 1:S45-50.

38. Boon M, Jorissen M, Jaspers M, Augustijns P, Vermeulen FL, Proesmans M, et al. The influence of nebulized drugs on nasal ciliary activity. J Aerosol Med Pulm Drug Deliv 2016;29:378-85.

39. Han LY, Wilson R, Slater S, Rutman A, Read RC, Snell NJ, et al. In vitro and in vivo effects of ribavirin on human respiratory epithelium. Thorax 1990;45:100-4.

40. Dolovich MB, Eng P, Mahony JB, Chambers C, Newhouse MT, Chernesky MA. Ciliary function, cell viability, and in vitro effect of ribavirin on nasal epithelial cells in acute rhinorrhea. Chest 1992;102:284-7.

41. Dimova S, Mugabowindelowe R, Willems T, Brewster ME, Noppe M, Ludwig A, et al. Safety-assessment of 3-methoxyquercetin as an antirhinoviral compound for nasal application: effect on ciliary beat frequency. Int J Pharm 2003;263:95-103.

42. Hofmann T, Reinisch S, Gerstenberger C, Koele W, Gugatschka M, Wolf G. Influence of topical antifungal drugs on ciliary beat frequency of human nasal mucosa: an in vitro study. Laryngoscope 2010;120:1444-8.

43. Jiao J, Zhang L. Effect of Amphotericin B on human nasal ciliary beat frequency. Zhonghua Er Bi Yan Hou Tou Jing Wai Ke Za Zhi 2016;51:573-7.

44. Cho DY, Hoffman KJ, Gill GS, Lim DJ, Skinner D, Mackey C, et al. Protective and antifungal properties of Nanodisk-Amphotericin B over commercially available Amphotericin B. World J Otorhinolaryngol Head Neck Surg 2017;3:2-8.

45. Jiang RS, Twu CW, Liang KL. Efficacy of nasal irrigation with 200 μg/ml amphotericin B after functional endoscopic sinus surgery: a randomized, placebo-controlled, double-blind study. Int Forum Allergy Rhinol 2018;8:41-8.

46. Uchenna Agu R, Jorissen M, Willems T, Van den Mooter G, Kinget R, Verbeke N, et al. Safety assessment of selected cyclodextrins - effect on ciliary activity using a human cell suspension culture model exhibiting in vitro ciliogenesis. Int J Pharm 2000;193:219-26.

47. Haffejee N, Du Plessis J, Müller DG, Schultz C, Kotzé AF, Goosen C. Intranasal toxicity of selected absorption enhancers. Pharmazie 2001;56:882-8.

48. Aspden TJ, Mason JD, Jones NS, Lowe J, Skaugrud O, Illum L. Chitosan as a nasal delivery system: the effect of chitosan solutions on in vitro and in vivo mucociliary transport rates in human turbinates and volunteers. J Pharm Sci 1997;86:509-13.

49. Bonengel S, Bernkop-Schnürch A. Thiomers--from bench to market. J Control Release 2014;195:120-9.

50. Palmberger TF, Augustijns P, Vetter A, Bernkop-Schnürch A. Safety assessment of thiolated polymers: effect on ciliary beat frequency in human nasal epithelial cells. Drug Dev Ind Pharm 2011;37:1455-62.
51. Mallants R, Jorissen M, Augustijns P. Effect of preservatives on ciliary beat frequency in human nasal epithelial cell culture: single versus multiple exposure. Int J Pharm 2007;338:64-9. 

52. Rizzo JA, Medeiros D, Silva AR, Sarinho E. Benzalkonium chloride and nasal mucociliary clearance: a randomized, placebo-controlled, crossover, double-blind trial. Am J Rhinol 2006;20:243-7.

53. Bernstein IL. Is the use of benzalkonium chloride as a preservative for nasal formulations a safety concern? A cautionary note based on compromised mucociliary transport. J Allergy Clin Immunol 2000;105:39-44.

54. Riechelmann H, Deutschle T, Stuhlmiiller A, Gronau S, Bürner H. Nasal toxicity of benzalkonium chloride. Am J Rhinol 2004;18:291-9.

55. Fokkens WJ, Lund VJ, Mullol J, Bachert C, Alobid I, Baroody F, et al. EPOS 2012: European position paper on rhinosinusitis and nasal polyps 2012. A summary for otorhinolaryngologists. Rhinology 2012;50:1-12.

56. Keojampa BK, Nguyen MH, Ryan MW. Effects of buffered saline solution on nasal mucociliary clearance and nasal airway patency. Otolaryngol Head Neck Surg 2004;131:679-82.

57. Roberts G, Katsikipisi M, Borrego LM, Custovic A, Halken S, Hellings PW, et al. Paediatric rhinitis: position paper of the European Academy of Allergy and Clinical Immunology. Allergy 2013;68:1102-16.

58. Bencova A, Vidan J, Rozborilova E, Kocan I. The impact of hypertonic saline inhalation on mucociliary clearance and nasal nitric oxide. J Physiol Pharmacol 2012;63:309-13.

59. Satdhabudha A, Poachanukoon O. Efficacy of buffered hypertonic saline nasal irrigation in children with symptomatic allergic rhinitis: a randomized double-blind study. Int J Pediatr Otorhinolaryngol 2012;76:583-8.

60. Hauptman G, Ryan MW. The effect of saline solutions on nasal patency and mucociliary clearance in rhinosinusitis patients. Otolaryngol Head Neck Surg 2007;137:815-21.

61. Michel O, Dreßler AK. Hypertonic (3%) vs. isotonic brine nosespray--a controlled study. Laryngorhinootologie 2011;90:206-10.

62. Ural A, Oktener TK, Kızıl Y, Ileri F, Uslu S. Impact of isotonic and hypertonic saline solutions on mucociliary activity in various nasal pathologies: clinical study. J Laryngol Otol 2009;123:517-21.

63. Boek WM, Keleş N, Graamans K, Huizing EH. Physiologic and hypertonic saline solutions impair ciliary activity in vitro. Laryngoscope 1999;109:396-9.

64. Min YG, Lee KS, Yun JB, Rhee CS, Rhyoo C, Koh YY, et al. Hypertonic saline decreases ciliary movement in human nasal epithelium in vitro. Otolaryngol Head Neck Surg 2001;124:313-6.

65. Ünal M, Göür K, Özcan C. Ringer-Lactate solution versus isotonic saline solution on mucociliary function after nasal septal surgery. J Laryngol Otol 2001;115:796-7.

66. Low TH, Woods CM, Ullah S, Carney AS. A double-blind randomized controlled trial of normal saline, lactated Ringer’s, and hypertonic saline nasal irrigation solution after endoscopic sinus surgery. Am J Rhinol Allergy 2014;28:225-31.

67. Süslü N, Bajin MD, Süslü AE, Oğretmenoğlu O. Effects of buffered 2.3%, buffered 0.9%, and non-buffered 0.9% irrigation solutions on nasal mucosa after septoplasty. Eur Arch Otorhinolaryngol 2009;266:685-9.

68. Fonuant S, Choiyasate S, Roongrongwatanasiri K. Comparison on the efficacy of dexpanthenol in seawater and saline in postoperative endoscopic sinus surgery. J Med Assoc Thai 2008;91:1588-63.
69. Bonnomet A, Luczka E, Coraux C, de Gabory L. Non-diluted seawater enhances nasal ciliary beat frequency and wound repair speed compared to diluted seawater and normal saline. Int Forum Allergy Rhinol 2016;6:1062-8.

PUBMED | CROSSREF

70. Laberko EL, Bogomil’sky MR, Soldatsky YL, Pogosova IE. The influence of an isotonic solution containing benzalkonium chloride and a hypertonic seawater solution on the function of ciliary epithelium from the nasal cavity in vitro. Vestn Otorinolaringol 2016;81:49-52.

PUBMED | CROSSREF