

REVIEW

Endothelin in nasal mucosa: role in nasal function and inflammation

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The endothelins (ETs) belong to a family of 21-amino acid peptides originally isolated from porcine aortic endothelial cells, which have potent vasoconstrictor, bronchoconstrictor and glandular secretory effects. ET has three genomic isoforms, ET-1, ET-2 and ET-3, which act through two receptor subtypes, ET_A and ET_B. ET is expressed in both the lower and upper respiratory mucosa in humans and several animal species. In human nasal mucosa, ET-1 mRNA and protein are widely expressed in epithelial and endothelial cells, submucosal glands, inflammatory cells, venous sinusoids and small arterioles. ET-1 binding sites have been located in submucosal glands, venous sinusoids, small arterioles, but not in the human nasal epithelium. In the nose, ET increases mucociliary activity in maxillary sinus, stimulates nasal glandular secretion and induces nasal symptoms. Since the expression of ETs is enhanced in glands and inflammatory cells in chronic inflammation, and ET-1 induces the secretion of pro-inflammatory mediators in human nasal mucosa, a role for ET in nasal polyposis and rhinitis has been suggested. Endothelin, in addition to eicosanoids and pro-inflammatory cytokines, would establish a pro-inflammatory loop that could become independent of the original stimulus, and contribute to long-term nasal mucosa inflammatory changes. ET-induced cytokine release could contribute to chemoattraction of pro-inflammatory cells such as eosinophils, neutrophils, B cells and T cells, which could in turn contribute to the perpetuation of nasal inflammation.

Introduction

Endothelins (ETs) constitute a family of 21-amino acid peptides originally isolated from the culture supernatant of porcine aortic endothelial cells [1], having potent vasoconstrictor, bronchoconstrictor and glandular secretory effects [2–7]. ETs have at least three genomic isoforms, ET-1, ET-2 and ET-3 [8], acting via two different receptors, ET_A and ET_B [9]. ET_A receptors bind ET-1 and ET-2 more avidly

than ET-3, while ET_B receptors bind all three with equal affinity. Since the discovery of ETs by Yanagisawa and coworkers [1], the role of ETs in the lower airways has been extensively studied and widely reviewed [2–7]. In contrast, very few studies in either animals or humans have investigated the role of ETs in nasal pathophysiology. The present review will focus its interest in the presence and the potential role of ETs in human nasal mucosa, in both health and disease.

ET immunoreactivity

In animals, ET was first reported to be expressed in the nasal mucosa of fetal rats [10] and the epithelium of rabbit maxillary sinus [11]. In human nasal mucosa, ET was first reported in the endothelial cells of both capacitance and resistance blood vessels [12]. Both immunoreactive ET-1 and endothelin-converting enzyme (ECE)-1 are expressed in epithelial and endothelial cells, submucosal glands (in mucous cells and to a greater extent in serous cells), inflammatory cells (macrophages), and blood vessels such as arterioles, small veins and venous sinusoids of human nasal mucosa [13–17] (Table 1). These findings contrast with the distribution of other peptides such as calcitonin gene-related peptide (CGRP), neuropeptide Y (NPY), gastrin-releasing peptide (GRP), vasoactive intestinal polypeptide (VIP), substance P (SP), and neurokinin A (NKA), which are mainly present in bundle nerves involved in adrenergic, cholinergic and nonadrenergic, noncholinergic functions [18–21]. Compared with healthy mucosa, the expression of ETs in glands and inflammatory cells is enhanced in nasal polyposis [15] and chronic rhinitis [16], suggesting a role of this molecule in nasal chronic inflammation.

In the lower airways, ETs are also produced and secreted by epithelial cells, macrophages, neuroendocrine cells, vascular smooth muscle, and glands of several animal species, including dogs, pigs, rats, mice, rabbits, and guinea pigs, as well as in humans [2–7]. Bronchial epithelial cell lines, such as rat L2 and human A549, are also able to release ET. Since stimuli such as thrombin, bacterial endotoxin, histamine and several cytokines increase its secretion in both animal and human airways, ET has been proposed as a regulatory

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Table 1. Expression of immunoreactive ET (irET), ET gene (mRNA) and ET receptors (ET_R) in human nasal mucosa

Tissues	irET	ET-1 mRNA	ET _R
Submucosal glands	+	+	+
Epithelial cells	+	+	–
Endothelial cells	+	+	+
Venous sinusoids	+	+	+
Arterioles	+	+	+
Small veins	+	+	+
Non-structural cells	+	+	–
References	13–17	13–17	13,17

+ = present, – = not present

peptide of bronchial and vascular tone and development, as well as in airway inflammation [2].

ET mRNA expression

In human nasal mucosa, both ET-1 and endothelin-converting enzyme (ECE)-1 mRNA are widely expressed in epithelial and endothelial cells, submucosal glands, inflammatory cells (macrophages), venous sinusoids and small arterioles [13–16] (Table 1). ET-1 mRNA has been detected by *in situ* hybridization in smooth muscle of venous sinusoids, small veins and small muscular arteries [13]. Some non-structural cells, possibly macrophages or mast cells [14,22], also express ET-1 mRNA. A recent study has reported ET-1 mRNA expression in epithelial cells and glands of human nasal mucosa with chronic inflammation [16]. Epithelial cells from both animal and humans lower airways as well as from bronchial epithelial cell lines, such as rat L2 and human A549 and BEAS-2B, also express ET mRNA [2,23].

ET receptors

ET-1 binding sites are found concentrated around submucosal glands, arterioles and venous sinusoids, but not in the epithelium of human nasal mucosa [13,17]. Although early studies done on animal lower airways reported ET-specific binding sites in the epithelium [10], no constitutive binding sites have been found in the human nasal epithelium.

In human lower airways, although early studies reported ET-1 receptors in tracheal smooth muscle and parasympathetic nerves but not in the epithelium, binding sites for ET-1, ET-2 and ET-3 have lately been found in the epithelium and the submucosa [2]. Several animal species such as rat, rabbit, cat and dog also express abundant ET receptors in the lower airways [2]. Recently, the use of specific receptor antagonists has identified two ET receptor subtypes, ET_A

and ET_B [4]. Low levels of ET_A, but not ET_B, receptors have been found in the bronchial epithelium of both asthmatic and nonasthmatic patients, and in epithelial cell lines such as rat L2 or human BEAS-2B [2]. In human nasal mucosa, the characterization and distribution of ET receptor subtypes (ET_A and ET_B) have recently been reported. ET_A receptors are localized in the vascular wall while ET_B receptors are mainly distributed in the endothelium [17].

Endothelin and eicosanoids

It has been suggested that some of the effects of ET in human nasal mucosa, such as the regulation of vascular tone [24] or the stimulation of glandular secretion [13], might be mediated, at least in part, by eicosanoid production. Under baseline conditions, human nasal mucosa predominantly generates cyclo oxygenase products with small amounts of lipooxygenase products [25]. Within one hour ET-1 induces the release of several prostanoids with a predominance of PGE₂, followed by PGD₂, PGF_{2α} and TXB₂. With a maximum effect at four hours, ET-1 also induced the production of 15-HETE [25]. No peptidoleukotriene release was found after incubation with culture media or ET-1. Although a study reported that ET-1-induced guinea pig airway constriction was attenuated by a PAF antagonist [26], Wu and colleagues found no changes in eicosanoid production after the addition of a potent PAF antagonist, Ro 19–3704 [25].

In both baseline and post-ET stimulation, cyclo oxygenase products dominated the amino acid metabolism cascade in human nasal mucosa, but any cell type could be responsible for this eicosanoid generation. PGE₂, the predominant metabolite in nasal explants, is known to induce bronchodilation, and inhibit lymphocyte immune function, fibroblast proliferation, macrophage cytokine production and cell differentiation [27,28]. PGD₂ causes peripheral vasodilation, pulmonary vasoconstriction, bronchoconstriction, airway hyperreactivity, and mucous secretion, while it inhibits platelet aggregation [29]. PGF_{2α} induces both vasoconstriction and bronchoconstriction, mucous secretion, and stimulates airway sensory nerves [30]. TXA₂ may induce vaso- and bronchoconstriction as well [25]. 15-HETE is released by both nasal and bronchial epithelium [31] and may induce mucous secretion and mitogenesis, and modulate immune functions and oxygenation enzymes [25]. Moreover, in allergic rhinitic patients nasal allergen challenge induces eicosanoid release, including 15-HETE [32]. Since 15-HETE is a potent eosinophil chemoattractant, this product could contribute to perpetuate airway inflammation.

In the lower airways of several animal species, such as rat, rabbit, dog and guinea pig, ET-1 also stimulates the release of eicosanoid products [2]. In cultured feline airway epithelial cells, ET-1 increases the release of PGE₂, PGF_{2α},

5-HETE and 15-HETE, acting through specific receptors [33]. In human bronchi, ET-1 also causes an increase in prostanoid release via ET_A receptors [2]. ET may elicit its biological responses by different signal transduction pathways, including the activation of G protein-coupled phospholipase C (PLC) and voltage-dependent calcium ion channels [34], as well as the activation of phospholipase A₂ (PLA₂) [35]. COX inhibitors have been reported to inhibit ET-induced mucociliary activity in human nasal mucosa [11]. In rat L2 epithelial cells, exogenous ET-1 increased cAMP and PGE₂ production, whereas blockade of endogenous ET-1 decreased PGE₂ production [2]. However, it is not yet clear whether ET-1 increases eicosanoid production only by activating PLA₂ and/or PLC or by stimulating cyclo oxygenases and lipoxygenases as well.

Endothelin and cytokines

Allergens, bacterial endotoxin and several cytokines, such as IL-1, IL-2, IL-6, IL-8, TNF α , TGF β , and IGF-1, have been reported to stimulate ET production in animal and human airway epithelial cells, as well as in epithelial cell lines [2]. Recent reports have also shown that anti-IgE stimulation induces ET release from asthmatic bronchial epithelial cells which express the low affinity receptor for IgE (Fc ϵ RII) [2]. This ET release may be responsible in part for the acute exacerbation and perpetuation of airway inflammation. Since no epithelial cell lines from either human or animal nasal mucosa are available, and the physiology of epithelial cells from both nasal and bronchial mucosa has been reported to be similar [36], immortalized bronchial epithelial cell lines have been used to study the effect of ET on epithelial cell cytokine release. BEAS-2B epithelial cells constitutively release higher concentrations of IL-8 followed by IL-6, G-CSF and GM-CSF [23,37]. Mullol *et al.* reported that cytokine release caused by ET-1 is biphasic with an early release (4 h) of IL-1 β , IL-6 and IL-8, and a late release (48 h) of IL-8 and GM-CSF [37]. Cytokine concentrations in BEAS-2B cells are similar to those the same authors have found in primary epithelial cells from both nasal mucosa and nasal polyps [38,39].

Since BEAS-2B cells express ET_A receptors and produce irET [23], it is possible that ET of epithelial and other origins may act in a paracrine–autocrine fashion to stimulate the production of IL-8, IL-6 and GM-CSF contributing to a relatively rapid inflammatory response to airway injury. Among the pro-inflammatory effects of these cytokines in airway inflammation, GM-CSF, IL-8 and TNF α are able to induce eosinophil survival and activation. These effects are abrogated by steroids, by both inhibiting epithelial cell cytokine release and causing eosinophil apoptosis [38–41].

In human umbilical cord endothelial cells (HUVEC), a combination of cytokines (TNF α , IL-1 β and IFN γ) and

monocyte chemotactic protein (MCP-1) also showed a potent effect on increasing the spontaneous expression of ET-1 mRNA and ET-1 peptide release [42].

ET functions

Nasal secretions

Mullol *et al.* reported that ET-1 and ET-2, but not ET-3, stimulate the release of glandular secretory products, such as lactoferrin and mucous glycoprotein, in explant cultures of human nasal mucosa [13]. Riccio *et al.* showed that ET-1 also induces nasal symptoms such as rhinorrhea, itching, and sneezing and glandular but not vascular secretion [43]. Nasal responses are more pronounced in allergic patients than in healthy subjects, suggesting that allergic inflammation might enhance nasal responsiveness to ET. Neutral endopeptidase (NEP), angiotensin-converting enzyme (ACE) and carboxypeptidases induce the degradation by hydrolysis of ETs and this effect is inhibited by protease inhibitors [13,44]. NEP and carboxypeptidases are produced and released in the nasal mucosa [45,46]. The incubation of nasal explants with a protease inhibitor, aprotinin, prevented ET-1 digestion by endogenous proteases, suggesting that proteases capable of reducing ET actions are present in nasal mucosa [13]. In addition, other events such as the release of epithelium-derived relaxant agents, may also modulate endothelin-induced responses in the nasal mucosa. Moreover, it is very interesting to note that ET, a highly potent vasoconstrictor, is also a potent secretagogue while other secretagogue peptides, such as VIP and SP [21,47], are vasodilators, clearly suggesting that glandular and vascular responses to these peptides may not be related. Since arterioles, venous sinusoids and submucosal glands possess ET-1 binding sites, it appears that ETs released from those and other sources may play a role in nasal physiology by acting upon submucosal glands to stimulate glandular secretion [13], in addition to the effects on arteriolar and venous sinusoids to regulate nasal blood flow [24]. In human bronchial mucosa, ET induces glandular product secretion as well [29].

Vascular tone

Although ET was reported to be a vasodilator peptide in pig airway mucosa [48], it induces a potent and prolonged vasoconstriction in nasal mucosa of pigs [49], dogs [50–52], rabbits [14,24,53], and humans [24,50,51]. This effect is calcium dependent [50,51] but is not prevented by COX inhibitors [24]. Furthermore, levels of ET-1 in nasal blood vessels are similar to those in the general blood vessels [54] (Table 2).

Table 2. Biological functions of endothelin in human nasal mucosa

Target tissue	Biological functions	References
Submucosal glands	Increase in glandular secretion	13,43
Epithelium	Not known (cytokine-eicosanoid release?)	(25)
Connective tissue	Not known	—
Vascular smooth muscle	Vasoconstriction	14,24,50,51
Mucociliary system	Increase in mucociliary activity	11
Whole nasal mucosa	Increase in PGE ₂ , PGD ₂ , PGF _{2α} , TXB ₂ , and 15-HETE release	25
	Increase in nasal symptoms (sneezing, nasal itching, obstruction, rhinorrhea)	43

Mucociliary activity

In the maxillary sinus of rabbits, the increase of mucociliary activity induced by ET and prevented by COX inhibitors also suggests a potential role for this molecule in both nasosinus homeostasis and inflammation [11].

Regulation of nasal physiology and inflammation

Although no ET binding sites have yet been located in human nasal epithelium [13], it has been reported that epithelial cells produce and release ETs, and express ET-specific binding sites and receptor mRNA. Locally released, ET-1 may participate in the regulation of nasal blood flow and secretory cell exocytosis via paracrine mechanisms [13,24]. Studies on animal and human nasal mucosa have shown that ET-1 induces a potent and prolonged vasoconstriction [14,24,53], suggesting that locally released ET-1, in association with other neurotransmitters, could participate in the control of nasal blood flow via a paracrine mechanism. All this and the fact that their circulating plasma concentrations are far below pharmacological levels [9], suggest that ETs may act as locally acting peptides (paracrine and autocrine) rather than circulating hormones. ET-1 intranasal administration induces nasal symptoms and glandular product release [43]. Similarly to the effect of bradykinin [2], nasal provocation with ET-1 also triggers nociceptive nerve activation, sneezing and parasympathetic glandular secretion, but no vascular permeability, in both allergic and nonallergic individuals [43]. These findings suggest that a nociceptive nerve-parasympathetic nervous system reflex activated by ET-1 is upregulated in allergic rhinitis, as well as providing evidence of a role for allergic inflammation in nasal mucosa hyperresponsiveness to ET-1. To support this data, there have also been reports of an increased co-expression of ET-1 and ECE-1 in human nasal mucosa from patients with chronic inflammation [16]. In contrast, a recent study has reported that ET-1 is present in the nasal epithelium of healthy subjects but absent in the metaplastic epithelium of chronic rhinitis [17].

Concluding remarks

The nasal mucosa acts as a physical and immunological barrier which protects the airways against inhaled pollutants and irritants. Total ET, released from epithelial and endothelial cells, glands and other sources may play an active role in nasal mucosa homeostasis by modulating epithelial permeability, vascular tone, glandular secretion, mucociliary activity, mediator release and inflammatory cell recruitment (Tables 1 and 2). The presence of specific receptors and the demonstration of total ET production in and by different cell types strengthen the hypothesis that this peptide may play an important role in nasal mucosa in both health and inflammation. It seems likely that ET (mainly ET-1) may function as a local hormone being elaborated by epithelial cells, glands and endothelial cells which acts on adjacent target tissues (Fig. 1). Nevertheless, the patterns of ET-2 and ET-3 production and expression in nasal mucosa have not been fully evaluated.

Extrapolations from experiments done in animals, human beings and epithelial cell lines clearly suggest that pro-inflammatory mediators such as cytokines, allergens or bacterial endotoxin may increase ET-1 production in conditions characterized by chronic airway inflammation, such as nasal polyposis, rhinitis or asthma. In consequence, total ET would cause a paracrine effect on several nasal target cells inducing glandular secretion, regulation of vascular tone and mucociliary activity, secretion of inflammatory mediators such as eicosanoids and cytokines, and pro-inflammatory cell chemotaxis, leading to the symptoms of rhinitis (sneezing, nasal itching and obstruction, and rhinorrhea).

In summary, endothelin and other pro-inflammatory mediators could establish a pro-inflammatory loop which, once initiated, could become independent of the original stimulus, and contribute to long-term inflammatory changes in the nasal respiratory mucosa. ET-induced cytokine release could contribute to chemoattraction of pro-inflammatory cells such as eosinophils, neutrophils, T lymphocytes and B lymphocytes, that could in turn contribute to the perpetuation of this inflammatory loop. Future animal and human

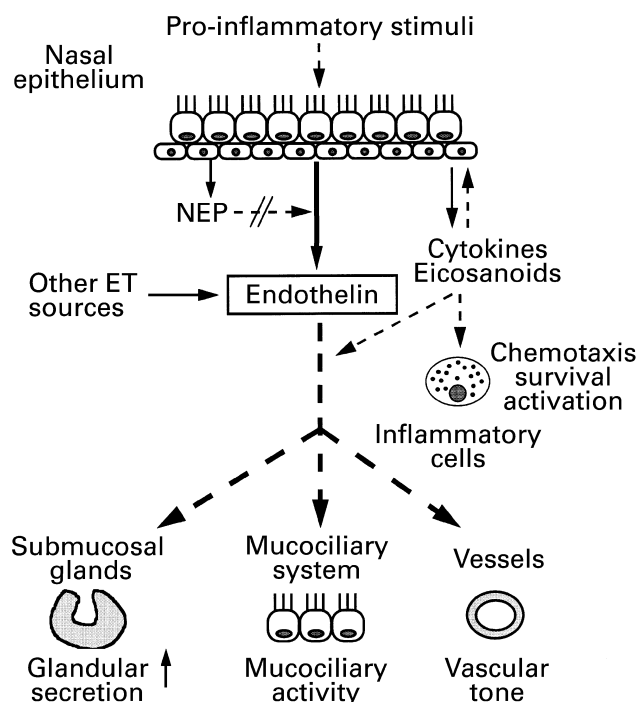


Fig. 1. The potential role of endothelin (ET) in healthy and inflamed human nasal mucosa. In a healthy situation, ET from epithelial cells and other sources would act, through a paracrine pathway, on nasal target tissues, such as submucosal glands, mucociliary system and blood vessels, to regulate mucous hypersecretion, mucociliary activity and vascular tone. Degradation by neutral endopeptidase (NEP) would regulate ET effects. During inflammation, pro-inflammatory stimuli such as endotoxin, allergens or cytokines would stimulate ET release from nasal epithelium, thus causing a pro-inflammatory loop and increasing its effects on nasal target tissues. Plain arrows = release, dashed arrows = effects

studies and the use of specific ET receptor antagonists will determine the real importance of this potential endothelin cascade in nasal inflammation.

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References

- 1 Yanagisawa M, Kurihara H, Kimura S et al. A novel potent vasoconstrictor peptide produced by vascular endothelial cells. *Nature* 1988; 332:411–5.
- 2 Mullol J, Baraniuk JN, Picado C, Shelhamer JH. Endothelin and the epithelium. In: Goldie RG, Hay DWP, eds. *Pulmonary Actions of the Endothelins*. Basel: Birkhäuser-Verlag AG, 1999:145–66.
- 3 Barnes PJ. Endothelins and pulmonary diseases. *J Appl Physiol* 1994; 77:1051–9.
- 4 Douglas SA, Meek TD, Ohlstein EH. Novel receptor antagonists welcome a new era in endothelin biology. *Tips* 1994; 15:313–6.
- 5 Filep JG. Endothelin peptides: biological actions and pathophysiological significance in the lung. *Life Sci* 1993; 52:119–33.
- 6 Hay DWP, Henry PJ, Goldie RG. Is endothelin-1 a mediator in asthma? *Am J Respir Crit Care Med* 1996; 154:1594–7.
- 7 Michael JR, Markewitz BA. Endothelins and the lung. *Am J Respir Crit Care Med* 1996; 154:555–81.
- 8 Inoue A, Yanagisawa M, Kimura S et al. The human endothelin family: three structurally and pharmacologically distinct isopeptides predicted by three separate genes. *Proc Natl Acad Sci USA* 1989; 86:2863–7.
- 9 Sakurai T, Yanagisawa M, Masaki T. Molecular characterization of endothelin receptors. *Tips* 1992; 13:103–7.
- 10 MacCumber MW, Ross CA, Snyder SH. Endothelin: visualization of mRNA by in situ hybridization provides evidence for local action. *Proc Natl Acad Sci USA* 1989; 86:7285–9.
- 11 Amble FR, Lindberg SO, McCaffrey TV, Runer T. Mucociliary function and endothelins 1, 2 and 3. *Otolaryngol Head Neck Surg* 1993; 109:634–45.
- 12 Casasco A, Galioto GB, Benazzo M et al. Presence of the regulatory peptide endothelin in the vascular endothelium of the human nasal mucosa. *Acta Otorhinolaryngol Ital* 1991; 11:185–8.
- 13 Mullol J, Chowdhury BA, White MV et al. Endothelin in human nasal mucosa. *Am J Respir Cell Mol Biol* 1993; 8:393–402.
- 14 Casasco A, Benazzo M, Casasco M et al. Occurrence, distribution and possible role of the regulatory peptide endothelin in the nasal mucosa. *Cell Tissue Res* 1993; 274:241–7.
- 15 Ohkubo K, Ohnishi M, Yokoshima K, Takizawa R, Okuda M, Kaliner MA. Study of endothelin: distribution in the airway and release from nasal epithelial cells. *Alerugi* 1994; 43:448–57.
- 16 Furukawa K, Saleh D, Bayan F et al. Co-expression of endothelin-1 and endothelin-converting enzyme-1 in patients with chronic rhinitis. *Am J Respir Cell Mol Biol* 1996; 14:248–53.
- 17 Gulisano M, Barni T, Fantoni G et al. Presence and location of endothelin and endothelin binding sites in human nasal mucosa under normal and metaplastic conditions. *Cell Tissue Res* 1998; 293:509–16.
- 18 Baraniuk JN, Lundgren JD, Goff J et al. Gastrin-releasing peptide in human nasal mucosa. *J Clin Invest* 1990; 85:998–1005.
- 19 Baraniuk JN, Castellino S, Lundgren JD et al. Neuropeptide Y (NPY) in human nasal mucosa. *Am J Respir Cell Mol Biol* 1990; 3:165–73.
- 20 Baraniuk JN, Lundgren JD, Goff J et al. Calcitonin gene-related peptide in human nasal mucosa. *Am J Physiol* 1990; 258:L81–8.

- 21 Baraniuk JN, Lundgren JD, Okayama M et al. Vasoactive intestinal peptide in human nasal mucosa. *J Clin Invest* 1990; 86:825–31.
- 22 Ehrenreich H, Anderson RW, Fox CH et al. Endothelins, peptides with potent vasoactive properties, are produced by human macrophages. *J Exp Med* 1990; 172:1741–8.
- 23 Mullol J, Baraniuk JN, Logun C, Picado C, Shelhamer JH. Endothelin-1 (ET-1) induces GM-CSF, IL-6 and IL-8 but not G-CSF release from a human bronchial epithelial cell line. *Am J Respir Crit Care Med* 1995; 151:A367.
- 24 Runer T, Lindberg S. Effects of endothelins 1, 2 and 3 on mucosal blood flow in the upper airways. *Acta Otolaryngol (Stockh)* 1996; 116:457–64.
- 25 Wu T, Mullol J, Rieves RD et al. Endothelin-1 stimulates eicosanoid production in cultured human nasal mucosa. *Am J Respir Cell Mol Biol* 1992; 6:168–74.
- 26 Battistini B, Sirois P, Braquet P, Filep J. Endothelin-induced constriction of guinea-pig airways: role of platelet-activating factor. *Eur J Pharmacol* 1990; 186:307–10.
- 27 Holtzman M. Arachidonic acid metabolism: implications of biological chemistry for lung function and disease. *Am Rev Respir Dis* 1991; 143:188–203.
- 28 Goodwin J. Immunomodulation by eicosanoids and anti-inflammatory drugs. *Curr Opin Immunol* 1989; 2:264–8.
- 29 Johnson CW, Rieves RD, Logun C et al. Endothelin-1 stimulates secretion of respiratory glycoprotein from human airways in vitro. *Am Rev Respir Dis* 1991; 143:A138.
- 30 Coleridge H, Coleridge J, Ginzel K, Baker D, Banzett R, Morrison M. Stimulation of irritant receptors and afferent C-fibers in the lungs by prostaglandins. *Nature* 1976; 264:451–3.
- 31 Henke D, Danilowicz R, Curtis J, Boucher R, Eling T. Metabolism of arachidonic acid by human nasal and bronchial epithelial cells. *Arch Biochem Biophys* 1988; 267:426–36.
- 32 Prat J, Mullol J, Ramis I et al. Release of chemical mediators and inflammatory cell influx during early allergic reaction in the nose. Effect of furosemide. *J Allergy Clin Immunol* 1993; 92:248–54.
- 33 Wu T, Rieves RD, Larivee P, Logun C, Lawrence MG, Shelhamer JH. Production of eicosanoids in response to endothelin-1 and identification of specific endothelin binding sites in airway epithelial cells. *Am J Respir Cell Mol Biol* 1993; 8:282–90.
- 34 Masaki T, Yanagisawa M, Takuwa Y, Yoshitoshi K, Kimura S, Goto K. Cellular mechanism of vasoconstriction induced by endothelin. In: Nishizuka Y, Endo M, Tanaka C, eds. *The Biology and Medicine of Signal Transduction* New York, NY: Raven Press, 1990:425–8.
- 35 Resink T, Scott-Burden T, Buhler F. Activation of phospholipase A₂ by endothelin in cultured vascular smooth muscle cells. *Biochem Biophys Res Commun* 1989; 158:279–86.
- 36 Devalia JL, Davies RJ. Human nasal and bronchial epithelial cells in culture: an overview of their characteristics and function. *Allergy Proc* 1991; 12:71–9.
- 37 Mullol J, Baraniuk JN, Logun C, Picado C, Shelhamer JH. Endothelin-1 (ET-1) induces GM-CSF, IL-6 and IL-8 but not G-CSF release from a human bronchial epithelial cell line (BEAS-2B). *Neuropeptides* 1996; 30:551–6.
- 38 Mullol J, Xaubet A, Gayà A et al. Cytokine gene expression and release from epithelial cells. A comparison study between healthy nasal mucosa and nasal polyps. *Clin Exp Allergy* 1995; 25:607–15.
- 39 Xaubet A, Mullol J, López E et al. Comparison of the role of nasal polyp and normal nasal mucosal epithelial cells on in vitro eosinophil survival. Mediation by GM-CSF and inhibition of dexamethasone. *Clin Exp Allergy* 1994; 24:307–17.
- 40 Roca-Ferrer J, Mullol J, López E et al. Effect of topical anti-inflammatory drugs on epithelial cell-induced eosinophil survival and GM-CSF secretion. *Eur Respir J* 1997; 10:1489–95.
- 41 Mullol J, López E, Roca-Ferrer J et al. Effects of topical anti-inflammatory drugs on eosinophil survival primed by epithelial cell. Additive effects of glucocorticoids and nedocromil sodium. *Clin Exp Allergy* 1997; 27:1432–41.
- 42 Baraniuk JN, Molet S, Mullol J, Naranch K. Endothelin and the airway mucosa. *Pulm Pharmacol* 1998; 11:113–23.
- 43 Riccio MM, Reynolds CJ, Hay DWP, Proud D. Effect of intranasal administrations of endothelin-1 to allergic and non-allergic individuals. *Am J Respir Crit Care Med* 1995; 152:1757–64.
- 44 Vijayaraghavan J, Scicli AG, Carretero OA, Slaughter C, Moomaw C, Hersh LB. The hydrolysis of endothelins by neutral endopeptidase 24.11 (enkephalinase). *J Biol Chem* 1990; 265:14150–5.
- 45 Ohkubo K, Baraniuk JN, Hohman RJ et al. Human nasal mucosal neutral endopeptidase (NEP): location, quantitation, and secretion. *Am J Respir Cell Mol Biol* 1993; 9:557–67.
- 46 Ohkubo K, Baraniuk JN, Mérida M, Hausfeld JN, Okada H, Kaliner MA. Human nasal mucosa carboxypeptidase: activity, location, and release. *J Allergy Clin Immunol* 1998; 96:924–31.
- 47 Baraniuk JN, Lundgren JD, Okayama M et al. Substance P and neurokinin A in human nasal mucosa. *Am J Respir Cell Mol Biol* 1991; 4:228–36.
- 48 Matran R, Alving K, Hemsén A, Lundberg JM. Endothelin-1 increases airway mucosa blood flow in the pig. *Agents Actions* 1990; 31(Suppl.):237–41.
- 49 Auberson S, Lacroix JS, Morel DR, Lehmann W, Lundberg JM. Methylene blue attenuates vasodilation and enhances vasoconstriction in response to endothelin-1 in the pig nasal mucosa. *Acta Physiol Scand* 1991; 142:149–56.
- 50 Ichimura K, Okita W, Tanaka T. Vasoactivity of endothelin in nasal blood vessels. *Rhinology* 1991; 29:125–35.
- 51 Okita W, Ichimura K, Tanaka T, Iinuma T. The vasoactivity of endothelin in the nasal mucosa. *Nip Jib Gak Ka* 1991; 94:291–9.
- 52 Okita W, Ichimura K, Tanaka T, Iinuma T. The effect of endothelin on neurotransmitter regulation of vascular tension in the nasal mucosa. *Nip Jib Gak Ka* 1992; 95:187–94.
- 53 Benazzo M, Casasco A, Lovotti P, Icaro Cornaglia A, Casasco M, Polak JM. Endothelin-induced vasoconstriction in rabbit nasal mucosa. *Acta Otolaryngol (Stockh)* 1994; 114:544–6.
- 54 Ichimura K, Okita W, Tanaka T. Plasma endothelin-1 concentration in the inferior turbinate. *J Otolaryngol* 1993; 22:83–5.