

Editorial

Transcriptional regulation of COX-2: a key mechanism in the pathogenesis of nasal polyposis in aspirin-sensitive asthmatics?

A. M. Vignola^{1,2}, V. Bellia¹

¹Istituto di Medicina Generale e Pneumologia, Unibersità di Palermo; ²Istituto di Biomedicina e Immunologia Molecolare – IBIM, CNR, Palermo, Italy

A. M. Vignola Istituto di Medicina Generale e Pneumologia Unibersità di Palermo, Italy Via Trabucco, 180 90146 Palermo, Italy

Accepted for publication 20 December 2002

The two isoforms of cyclooxygenase (known as COX-1 and COX-2) catalyze the initial step in the formation of biologically important prostanoids, such as prostaglandin (PG) E₂ and thromboxane (TX) A₂, in a variety of pathophysiologic processes. These include modulation of the inflammatory reaction, gastrointestinal cytoprotection and ulceration, angiogenesis and cancer, hemostasis and thrombosis. The PGs not only play a central role in inflammation, but also regulate other critical physiologic responses, including blood clotting, bone metabolism, wound healing, kidney function, blood vessel tone, and immune responses. PGs are usually synthesized in a broad range of tissue types and serve as autocrine or paracrine mediators to signal changes within the immediate environment.

The lungs are an important site of PGE₂ production and its immunoregulatory effects have been studied extensively in an animal model of ovalbumin-induced bronchial hyperreactivity. In mice with either COX-1 or COX-2 knockout, an increased broncho-alveolar fluid eosinophilia accompanied by macrophage and lymphocyte influx following ovalbumin challenge has been described (1), pointing out the important role that these enzymes play in the control of the inflammatory response into the airways.

COX-1 and COX-2 can also play an important role in aspirin-sensitive rhinitis and asthma. Although this phenomenon is not mediated by allergic mechanisms, it is known that specific COX-2 inhibitors are well tolerated by aspirin-sensitive asthmatic patients and do not trigger Cys-leukotrienes and PGD₂ release (2). In addition, asthmatic attacks are precipitated by COX-1 inhibition in aspirin-sensitive subjects, and recent findings indicate

alterations in basal arachidonic acid metabolism (3). Indeed, it has been found that, in contrast to atopic patients (4), a diminished expression of COX-2 is present in nasal mucosa of aspirin-intolerant patients (5) supporting the concept that COX-1 and COX-2 may function as a biologic brake in response to NSAIDs through the release of PGE₂ (6).

In this issue of the Journal, the data reported by Picado et al. show that the mean levels of COX-2 mRNA expression in nasal polyps from aspirin hypersensitivity asthma or rhinitis (AIAR) subjects is significantly lower than in subjects without aspirin hypersensitivity asthma or rhinitis (ATAR). In addition, they provide evidence that COX-2 downregulation is associated with a decreased expression and activation of NF- κ B, a transcription factor which modulates the activation of several genes, including that of COX-2. This finding is of great interest as it may provide a mechanistic explanation of the reduced COX-2 expression in the nasal mucosa of aspirin-sensitive subjects, and highlight for the first time the potential involvement of the NF- κ B system in the pathogenesis of chronic rhinosinusitis with nasal polyposis in aspirin-sensitive asthmatics.

The transcription factor NF- κ B plays a central role in immune and inflammatory responses. Indeed, it induces expression of many inflammatory mediators and is itself activated by inflammatory stimuli (7). NF- κ B is a dimer comprising subunits that can include c-Rel, RelA (p65), RelB, p50 and p52. In most cells, the NF- κ B prototype is a heterodimer composed of p65 and p50, the former subunit carrying the transactivating function (8). Endogenous inhibitors, known as I κ B, tightly regulate NF- κ B

activation by complexing with the transcription factor and trapping it in the cytoplasm. I κ B molecules form a distinct family of proteins that include I κ B α , I κ B β , I κ B ϵ , I κ B δ , Bcl3, p105 (9). The most characterized NF- κ B inhibitor is I κ B α . This protein binds avidly to the p65 subunit of NF- κ B through its ankyrin repeat domains that associates with the nuclear localization signal and the Ig-like domain of p65 (10).

During activation of NF- κ B, numerous stimuli, including IL-1 and TNF- α , activate a complex of I κ B kinases (IKK) that phosphorylate I κ B α on the amino terminus at serine residues 32 and 36 (11). The I κ B kinase complex contains two catalytic subunits, IKK α and IKK β , both of which phosphorylate I κ B, and a regulatory subunit, IKK γ , which is postulated to serve as a recognition site on I κ B α . Phosphorylation of I κ B α leads to its immediate polyubiquitinylation which targets I κ B α for rapid degradation by the proteasome. As a result, free NF- κ B dimers translocate to the nucleus and activate transcription of target genes [reviewed in reference (9)]. NF- κ B is clearly one of the most important regulators of the expression of several cytokines, such as TNF- α , IL-1 β , IL-6, and IL-8.

Interestingly, NF- κ B is also able to modulate the expression of COX-2 and therefore, as hypothesized in the paper by Picado et al., it may be involved in the abnormal expression of COX-2 in aspirin-sensitive asthmatic subjects. The evidence that both the expression and the activation of NF- κ B are reduced in AIAR than in ATAR supports this hypothesis and reiterates the concept that COX-2 downregulation in nasal polyps of AIAR subjects may represent the end-result of a complex, multistep process involving transcriptional and/or transductional pathways mediating the cellular responses to the external microenvironment. There are, however, a number of unexplored issues that still need to be addressed.

The first is related with the mechanisms underlying inflammation in rhinosinusitis in aspirin-sensitive subjects and the key role played by the activation of the NF- κ B system in its development. Indeed, it is well established that nasal polyps are characterized by a chronic eosinophilic inflammation as a result of an increased migration, activation, and survival of eosinophils into tissue (12). These processes are sustained by several cytokines, such as IL-3, IL-5 and GM-CSF, whose release is strictly dependent on the activation of the NF- κ B pathway (13, 14). The evidence provided by Picado et al. show that although the nasal mucosa of patients with and without aspirin hypersensitivity have the same pattern of NF- κ B subunit composition, its activity is significantly lower in patients with aspirin hypersensitivity. This evidence raises the issue that the molecular mechanisms regulating the inflammatory process underlying nasal polyposis are different in the two groups of subjects studied. The limit of this finding, however, lies in its descriptive nature, as no data are provided as to whether NF- κ B expression and/or activity is a cause or simply an epiphenomenon of

COX-2 downregulation. In addition, NF- κ B may not be the unique pathway involved in the modulation of the expression of COX-2, but other factors, such as MAP kinase (15), AP-1 (16) and nuclear factor IL-6 (17) may play a role. Furthermore, it has also been demonstrated that COX-2 and PGE-2 in pulmonary A549 cells does not involve activation of NF- κ B (18), indicating the complexity of the transcriptional regulation of COX-2 expression. Thus, the link between changes of NF- κ B activity and COX-2 expression is still not fully clear; in addition, provided that NF- κ B activation is reduced in AIAR, it still remains to be elucidated what intracellular signaling pathways are involved in the production of the wide range of inflammatory mediators that are continuously released in the nasal mucosa in AIAR subjects.

The second issue that deserves more studies is whether the reduced NF- κ B expression and activation occur similarly in the different cell populations of the nasal mucosa and of nasal polyps of AIAR subjects. In particular, it is extremely useful to understand whether the reduced NF- κ B activation takes place mainly in structural cells (i.e. epithelial cells and fibroblasts) and/or mobile cells (i.e. eosinophils, lymphocytes). Picado et al. assess NF- κ B expression by RT-PCR of total RNA from nasal mucosa and nasal polyps but do not provide evidence about its cellular and subcellular localization precluding the possibility to differentiate the NF- κ B activation in distinct cell types located in nasal polyps. It also remains to be established whether or not NF- κ B and COX-2 downregulation occurs concomitantly in the same cell types, a finding that would further strengthen the relationship between the two phenomena.

The third important issue deals with the pleotropic activity of COX-2 and with the need to better define its pathophysiologic role in the pathogenesis of nasal polyposis in AIAR subjects. Previous reports showed an enhanced expression of COX-2 in the airways of asthmatics with (19) or without aspirin sensitivity (20) and suggested that COX-2-derived metabolites may play an essential role in airway inflammation. In addition, in normal subjects, it has been shown that rhinovirus colds can also induce bronchial inflammation with markedly enhanced expression of COX-2 (21). Taken all together, these evidence suggest: (1) that COX-2 expression in aspirin-sensitive asthmatics is not solely localized in the nasal mucosa but can also be observed in the lower airways. In addition, the modulation of its expression appears to differ in the bronchi and in the nose of aspirin-sensitive subjects pointing out potential differences of the mechanisms regulating COX-2 expression at both sites. Moreover, whether or not this may be considered a systemic feature of AIAR is a fascinating point that needs to be better clarified; (2) that COX-2 overexpression may not necessarily lead to a protection against inflammation. Indeed, COX-2 can indeed promote cell survival (22), can stimulate angiogenesis (23), and can promote Th-2 type immune responses (24) suggesting that any strategy

targeted at the upregulation of COX-2 expression or NF- κ B activation deserves a great caution.

So far, investigations into the role of COX-2 in disease have suggested that chronic activation of this enzyme may be pathologic and that its inhibition may even limit cancer progression (24). Therefore, future studies into the function of COX-2 and on the regula-

tion of mechanisms regulating COX-2 gene activation and transcription represent an urgent need to better understanding the pathogenesis of aspirin-sensitive asthma. This will probably lead to new approaches for the treatment and, perhaps, prevention of immune disorders associated with inflammation and with aspirin hypersensitivity.

References

1. GAVETT SH, MADISON SL, CHULADA PC et al. Allergic lung responses are increased in prostaglandin H synthase-deficient mice. *J Clin Invest* 1999;**104**:721–732.
2. SZCZEKLIK A, NIZANKOWSKA E, BOCHENEK G, NAGRABA K, MEJZA F, SWIERCZYNSKA M. Safety of a specific COX-2 inhibitor in aspirin-induced asthma. *Clin Exp Allergy* 2001;**31**:219–225.
3. KOWALSKI ML, PAWLICZAK R, WOZNIAK J et al. Differential metabolism of arachidonic acid in nasal polyp epithelial cells cultured from aspirin-sensitive and aspirin-tolerant patients. *Am J Respir Crit Care Med* 2000;**161** (2 Pt 1):391–398.
4. GRZEGORCZYK J, KOWALSKI ML, KORNATOWSKI T, PAWLICZAK R. Expression of cyclooxygenase in nasal polyps from atopic and nonatopic subjects. *J Invest Allergol Clin Immunol* 1999;**9**:380–385.
5. PICADO C, FERNANDEZ-MORATA JC, JUAN M et al. Cyclooxygenase-2 mRNA is downexpressed in nasal polyps from aspirin-sensitive asthmatics. *Am J Respir Crit Care Med* 1999;**160**:291–296.
6. SZCZEKLIK A. Prostaglandin E2 and aspirin-induced asthma. *Lancet* 1995;**345**:1056.
7. RAHMAN I, MACNEE W. Role of transcription factors in inflammatory lung diseases. *Thorax* 1998;**53**:601–612.
8. SCHMITZ ML, BAEUERLE PA. The p65 subunit is responsible for the strong transcription activating potential of NF- κ B. *Embo J* 1991;**10**:3805–3817.
9. ROTHWART DM, KARIN M. The NF- κ B activation pathway: a paradigm in information transfer from membrane to nucleus. *Sci STKE* 1999;**1999**:1122–1130.
10. BAEUERLE PA, HENKEL T. Function and activation of NF- κ B in the immune system. *Annu Rev Immunol* 1994;**12**:141–179.
11. TRAENCKNER EB, PAHL HL, HENKEL T, SCHMIDT KN, WILK S, BAEUERLE PA. Phosphorylation of human I kappa B-alpha on serines 32 and 36 controls I kappa B-alpha proteolysis and NF- κ B activation in response to diverse stimuli. *Embo J* 1995;**14**:2876–2883.
12. SZCZEKLIK A, NIZANKOWSKA E, SANAK M, SWIERCZYNSKA M. Aspirin-induced rhinitis and asthma. *Curr Opin Allergy Clin Immunol* 2001;**1**:27–33.
13. TAKENO S, HIRAKAWA K, UEDA T, FURUKIDO K, OSADA R, YAJIN K. Nuclear factor-kappa B activation in the nasal polyp epithelium: relationship to local cytokine gene expression. *Laryngoscope* 2002;**112**:53–58.
14. RAMIS I, BIOQUE G, LORENTE J et al. Constitutive nuclear factor-kappaB activity in human upper airway tissues and nasal epithelial cells. *Eur Respir J* 2000;**15**:582–589.
15. SCHERLE PA, JONES EA, FAVATA MF et al. Inhibition of MAP kinase kinase prevents cytokine and prostaglandin E2 production in lipopolysaccharide-stimulated monocytes. *J Immunol* 1998;**161**:5681–5686.
16. OKADA Y, VOZNESENSKY O, HERSCHMAN H, HARRISON J, PILBEAM C. Identification of multiple cis-acting elements mediating the induction of prostaglandin G/H synthase-2 by phorbol ester in murine osteoblastic cells. *J Cell Biochem* 2000;**78**:197–209.
17. YAMAMOTO K, ARAKAWA T, UEDA N, YAMAMOTO S. Transcriptional roles of nuclear factor kappa B and nuclear factor-interleukin-6 in the tumor necrosis factor alpha-dependent induction of cyclooxygenase-2 in MC3T3-E1 cells. *J Biol Chem* 1995;**270**:31 315–31 320.
18. NEWTON R, HART L, CHUNG KF, BARNES PJ. Ceramide induction of COX-2 and PGE(2) in pulmonary A549 cells does not involve activation of NF- κ B. *Biochem Biophys Res Commun* 2000;**277**:675–679.
19. SOUSA A, PFISTER R, CHRISTIE PE et al. Enhanced expression of cyclo-oxygenase isoenzyme 2 (COX-2) in asthmatic airways and its cellular distribution in aspirin-sensitive asthma. *Thorax* 1997;**52**:940–945.
20. REDINGTON AE, MENG QH, SPRINGALL DR et al. Increased expression of inducible nitric oxide synthase and cyclo-oxygenase-2 in the airway epithelium of asthmatic subjects and regulation by corticosteroid treatment. *Thorax* 2001;**56**:351–357.
21. SEYMOUR ML, GILBY N, BARDIN PG et al. Rhinovirus infection increases 5-lipoxygenase and cyclooxygenase-2 in bronchial biopsy specimens from nonatopic subjects. *J Infect Dis* 2002;**185**:540–544.
22. BRUNE B, VON KNETHEN A. The role of nitric oxide and cyclooxygenase-2 in attenuating apoptosis. *J Environ Pathol Toxicol Oncol* 2002;**21**:103–112.
23. GATELY S. The contributions of cyclo-oxygenase-2 to tumor angiogenesis. *Cancer Metastasis Rev* 2000;**19**:19–27.
24. HARRIS SG, PADILLA J, KOUMAS L, RAY D, PHIPPS RP. Prostaglandins as modulators of immunity. *Trends Immunol* 2002;**23**:144–150.