Review Article

The roles of prostanoids in inflammation, allergy, and immunity

Toshiyuki Matsuoka, and Shuh Narumiya *
Department of Pharmacology, Faculty of Medicine, Kyoto University, Kyoto, Japan

Prostanoids, consisting of the prostaglandins (PGs) and the thromboxanes (TXs), are a group of lipid mediators formed in response to various stimuli. They include PGD₂, PGE₂, PGF₂α, PGI₂, and TXA₂. Given that aspirin-like nonsteroidal anti-inflammatory drugs exert their actions by suppressing prostanoid production, prostanoids have been implicated in processes inhibited by these drugs, including inflammation, fever, and pain. However, how prostanoids exert such a variety of actions had remained unclear. Prostanoids are released outside of the cells immediately after synthesis, and exert their actions by binding to receptors on the surface of target cells. We have identified a family of eight types or subtypes of G protein-coupled receptors that mediate prostanoid actions. They are the PGD receptor (DP), four subtypes of the PGE receptor (EP₁, EP₂, EP₃, and EP₄), the PGF receptor (FP), PGI receptor (IP), and TXA receptor (TP). Recently mice deficient in each of these prostanoid receptors were generated and subjected to various experimental models of disease. These studies have not only elucidated the molecular and cellular mechanisms of known prostanoid actions but also revealed previously unknown actions. In this article, we review several recent findings of the roles of prostanoid receptor signalling in inflammation, allergy, and immunity.

Rec./Acc.8/27/2008, pp423-433

* Correspondence should be addressed to:
Shuh Narumiya, Department of Pharmacology, Faculty of Medicine, Kyoto University, Kyoto 606-8501, Japan. Phone: +81-75-753-4392, Fax: +81-75-753-4693, E-mail: snaru@four.med.kyoto-u.ac.jp

Key prostanoid, receptor, inflammation, allergy, immunity

Introduction

Prostanoids are lipid mediators that modulate various physiological and pathophysiological functions throughout our body1-2. They are synthesized by sequential metabolism of arachidonic acid (AA), a C20 unsaturated fatty acid normally esterified to the sn-2 position of membrane glycerophospholipids. Prostanoids are produced “on demand”, rather than released from stores in the cell of origin, and act primarily as autacoids on the parent and/or neighboring cells with very short biological half-lives to mediate a wide variety of cellular interactions in physiological and pathological processes being involved in hemostasis/thrombosis, glomerular filtration and water balance, ovulation, embryo implantation and development, initiation of labor/abortion, inflammation and modulation of immunological responses. However, the mechanisms underlying such actions had remained a mystery until the receptors responsible were identified, their cDNAs cloned, and their roles analyzed. In this review, we review several recent findings of
the specific involvement of prostanoid receptor signalling in inflammation, allergy, and immunity.

**Biosynthesis of Prostanoids**

When tissues are exposed to diverse physiological and pathological stimuli, AA is liberated from membrane phospholipids and is converted to prostanoids, including the prostaglandins (PGs) and the thromboxanes (TXs) (Fig. 1). The cyclooxygenase (COX) reaction results in the formation of an unstable endoperoxide intermediate, PGG₂, which, in turn, is metabolized to PGD₂, PGE₂, PGF₂α, PGI₂, and TXA₂ by cell-specific synthases. The type of prostanoid produced by a given cell largely depends on the expression profile of the individual prostaglandin synthases. The physiological importance of prostaglandins is highlighted by the use of the COX-inhibiting nonsteroidal anti-inflammatory drugs (NSAIDs) in the clinical treatment of various disorders.

There are two distinct isoforms of COX encoded by separate genes, COX-1 and COX-2⁵. The two COX isoforms from the same species are 60–65% identical in the amino acid sequence, and catalyze the reaction in a mechanistically similar fashion with about the same Km values for AA. However, they differ in their expression and regulation⁶. While COX-1 is constitutively expressed in most tissues, COX-2 is virtually undetectable under the resting conditions and induced dramatically in response to various physiological and pathological stimuli. Therefore, prostanoids produced via COX-1 are usually believed to function in physiological homeostasis, while those generated via COX-2 are responsible for the inflammatory effects⁶. COX-2 gene expression has been shown to be under the control of the nuclear factor-kappa B (NF-κB)/Rel transcription factor family in response to pro-inflammatory stimuli such as lipopolysaccharide (LPS), interleukin (IL)-1, tumor necrosis factor (TNF)-α⁷. This is worthy of note because this group of transcription factors plays a central role in the immune response as well⁷. In experimental animals, COX-2 induction in response to inflammatory cytokines and stimuli such as bacterial LPS has been analyzed extensively for their high clinical relevance. Because traditional NSAIDs have deleterious side effects such as gastrointestinal bleeding due to suppression of both COX-1 and COX-2, COX-2-selective inhibitors were developed for anti-inflammatory action without deleterious side effects⁸. Although COX-2-selective inhibitors have become widely used for their benefits of reducing inflammatory actions and sparing constitutive actions of prostanoids, clinical trials revealed increased risks of cardiovascular adverse events, such as heart attacks and strokes, suggesting a cautious use of it⁹. Notably, the expression of COX-2, but not COX-1, is suppressed by glucocorticoids such as dexamethasone.

The conversion of PGH₂ to PGD₂ is catalyzed by PGD synthase (PGDS)¹⁰. There are two distinct types of PGDS: one is lipocalin-type PGDS localized in the central nervous system, male genitals, and heart; and the other is hematopoietic PGDS in mast cells and T helper type 2 (Th2) lymphocytes. Similarly, conversion of PGH₂ to PGE₂ is catalyzed by PGE synthases. To date, at least three members are known, namely microsomal PGES (mPGES)-1, mPGES-2 and cytosolic PGES (cPGES)¹¹. mPGES-1 is a perinuclear protein belonging to the MAPEG (membrane-associated proteins involved in eicosanoid and GSH metabolism) family. Like COX-2, this enzyme is markedly induced by proinflammatory stimuli, and down-regulated by anti-inflammatory glucocorticoids. Thus, mPGES-1 is thought to be functionally coupled with COX-2 in marked preference to COX-1. mPGES-2 rather constitutively expressed in various cells is synthesized as a Golgi membrane-associated protein, and the proteolytic removal of the N-terminal hydrophobic domain leads to the formation of a mature cytosolic enzyme. cPGES is also constitutively expressed in a wide variety of cells and is thought to be functionally linked to COX-1. Conversion of PGH₂ to PGF₂α is performed by the action of PGF synthase, that is a cytosolic enzyme belonging to the aldo-keto reductase family¹².
Conversion of PGI₂ to TXA₂ and PGI₃ is catalyzed by TX synthase and PGI synthase, respectively, both of which are heme-proteins belonging to the cytochrome P-450 family and are reportedly localized in the endoplasmic reticulum and perinuclear membranes

Prostanoid Receptors

Prostanoids thus formed are released outside of the cells immediately after synthesis. Because they are either chemically or metabolically unstable, it is believed that prostanoids work only locally, near their site of production. Prostanoids exert a variety of actions in various tissues and cells via membrane receptors on the surface of target cells. A family of membrane receptors mediating their actions has been characterized and cloned.

There are eight types and subtypes of receptors for prostanoid that are conserved in mammals from mouse to human: the PGD receptor (DP), four subtypes of the PGE receptor (EP₁, EP₂, EP₃, and EP₄), the PGF receptor (FP), the PGI receptor (IP), and the TXA receptor (TP). All are G protein-coupled rhodopsin-type receptors with seven transmembrane domains (Fig. 2A), and each is encoded by different genes. Despite the presence of conserved sequences, the overall homology among prostanoid receptors is not high, ranging from approximately 20 to 30 %.

However, the homology among receptor homologues from various species is considerably higher. The amino acids and nucleotides sequence homology between human and mouse DP, EP₁, EP₂, EP₃, EP₄, FP, IP and TP is 73, 84, 88, 84, 88, 79 and 76%, respectively. In addition, there are several splice variants of the EP₁, FP, and TP receptors, which differ only in their C-terminal tails. Thus far, three isoforms of the mouse EP₃ and eight of the human EP₁ have been identified, two of the human TP have been cloned, and for the FP two from sheep. Eight types of the prostanoid receptors can be grouped into three by their intracellular signalings (Fig. 2B). The IP, DP, EP₂, and EP₄ receptors mediate a cAMP rise and have been termed “relaxant” receptors, whereas the TP, FP, and EP₁ receptors induce calcium mobilization and constitute a “contractile” receptor group.

The EP₃ receptor induces a decline in cAMP levels and has been termed the “inhibitory” receptor. However, the effects of prostanoids on these G protein-coupled signalling pathways may change as a function of ligand concentration or structure. In addition to this prostanoid receptor families, there is a distinct type of PGD receptor, chemoattractant receptor-homologous molecule expressed on Th2 cells (CRTH₂). The CRTH₂ receptor belongs to the family of chemokine receptors, and mediates chemotaxis to PGD₂ of Th2 lymphocytes as well as eosinophils or basophils.

The roles of PGs and their receptor signalling in various physiological and pathophysiological conditions have been examined by comparing the effects of aspirin-like drugs with those of each prostanoid added exogenously. However, such studies do not clearly indicate which type of prostanoid and which class of prostanoid receptor is involved in a given process, nor how critical the actions of prostanoids might be. To address these questions, mice deficient in each prostanoid receptor have been generated and analyzed. In addition, highly selective agonists and antagonists for the cloned receptors have been developed (Table 1). Examinations of these mice combined with selective compounds were then performed and the important roles of each PG receptor signaling under various physiological and pathological conditions were revealed (Table 2). Among these
Table 1  Binding specificity of prostanoid analogs

<table>
<thead>
<tr>
<th>Compounds before the cloning of the prostanoid receptors</th>
<th>DP</th>
<th>EP1</th>
<th>EP2</th>
<th>EP3</th>
<th>EP4</th>
<th>FP</th>
<th>IP</th>
<th>TP</th>
</tr>
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<tbody>
<tr>
<td>Illoprost</td>
<td>&gt;10000</td>
<td>21</td>
<td>1600</td>
<td>22</td>
<td>2500</td>
<td>&gt;10000</td>
<td>11</td>
<td>&gt;10000</td>
</tr>
<tr>
<td>Carbachexin</td>
<td>&gt;10000</td>
<td>&gt;10000</td>
<td>1600</td>
<td>31</td>
<td>2300</td>
<td>1200</td>
<td>110</td>
<td>&gt;10000</td>
</tr>
<tr>
<td>TP-phenyl-PE2</td>
<td>&gt;10000</td>
<td>14</td>
<td>&gt;10000</td>
<td>4</td>
<td>1600</td>
<td>60</td>
<td>&gt;10000</td>
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<tr>
<td>L-644388</td>
<td>0.3</td>
<td>25400</td>
<td>267</td>
<td>3730</td>
<td>9250</td>
</tr>
<tr>
<td>ONO-8715</td>
<td>&gt;10000</td>
<td>0.3</td>
<td>3000</td>
<td>1000</td>
<td>&gt;10000</td>
</tr>
<tr>
<td>ONO-AE1-259</td>
<td>&gt;10000</td>
<td>&gt;10000</td>
<td>3</td>
<td>&gt;10000</td>
<td>&gt;10000</td>
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<tr>
<td>ONO-AE-248</td>
<td>&gt;10000</td>
<td>&gt;10000</td>
<td>3700</td>
<td>6</td>
<td>&gt;10000</td>
</tr>
<tr>
<td>ONO-AE-329</td>
<td>&gt;10000</td>
<td>&gt;10000</td>
<td>2100</td>
<td>1200</td>
<td>10</td>
</tr>
</tbody>
</table>

Dissociation constants (nM) for binding of each compound to the eight types or subtypes of mouse prostanoid receptors are shown.

Table 2  Major phenotypes of mice deficient in prostanoid receptors

<table>
<thead>
<tr>
<th>Phenotypes</th>
<th>EP1*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Decreased allergic responses in ovalbumin-induced bronchial asthma</td>
<td>Impaired PGD2-induced sleep</td>
</tr>
<tr>
<td>Decreased defensive D1 and D2 receptor signaling in the sinuses</td>
<td>Impaired TH1 differentiation of T lymphocyte</td>
</tr>
<tr>
<td>Decreased nonantigen specific antibody responses</td>
<td>Decreased collagen-induced arthritis</td>
</tr>
<tr>
<td>Decreased inflammatory gene expression</td>
<td>Decreased amyloid-beta formation in Alzheimer’s disease</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>Phenotypes</th>
<th>EP2*</th>
</tr>
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<tbody>
<tr>
<td>Impaired feeding in response to pyrogens</td>
<td>Impaired stress responses (ACTH release and stress behaviors)</td>
</tr>
<tr>
<td>Impaired arthritis</td>
<td>Impaired complex of inflammation</td>
</tr>
<tr>
<td>Decreased collagen-induced arthritis</td>
<td>Decreased collagen-induced arthritis</td>
</tr>
<tr>
<td>Decreased collagen-induced arthritis</td>
<td>Decreased target gene expression</td>
</tr>
</tbody>
</table>

Fever Generation

Infectious diseases activate the host immune system and raise inflammatory reactions against invaders. These responses cause general and characteristic central symptoms in patients, so-called sickness behaviors, including fever generation, adrenocorticotropic hormone release, reduced locomotion, etc. Fever is one of the representative symptoms of the sickness behaviors, which improve survival during bacterial infection. Both cellular components of infectious organisms and noninfectious inflammatory insults stimulate the production of cytokines such as IL-1, IL-6, and TNF-α, that then work as endogenous pyrogens and stimulates the neural pathways to raise body temperature. Given the very low possibility that cytokines can cross the blood-brain barrier, it is suggested that fever generation is mediated by small molecules which can diffuse into brain parenchyma through an immune system-brain interaction. Fever can be suppressed by NSAIDs, which indicates that PGs are important in fever generation. In addition, it is also reported that systemic administration of LPS induces COX-2 and mPGES expression in endothelial cells of brain microvessels in the organum vasculosum laminae terminalis (OVLT), a presumed site of pyrogen action. The roles of COX-2 and mPGES-1 in fever generation were then examined by the genetic deletion of these enzymes, and the essential role of COX-2 and mPGES-1 has been demonstrated by loss of fever generation to LPS in mice lacking COX-2 or mPGES-1.
Inflammatory Tachycardia

Systemic infection induces various adaptive responses including tachycardia. Although inflammation-associated tachycardia has been thought to result from increased sympathetic discharge caused by inflammatory signals of the immune system, definitive proof has been lacking. Using mice deficient in prostanoid receptors, Takayama et al. addressed the involvement of prostanoids in this process\(^{34}\). Administration of LPS to WT mice induced a biphasic increase in heart rate characterized by a transient peak (early phase) at 20 min followed by a sustained increase (late phase) that persisted for at least 100 min after LPS injection (Fig. 4A). Contribution of COX-1 and COX-2 was examined by using their selective inhibitors, SC560 and SC58125, respectively. SC560 suppressed the early phase, and SC58125 suppressed the late phase of LPS-induced tachycardia, indicating that COX-1 and COX-2 were involved preferentially in the early and late phases, respectively (Fig. 4A). In TP-deficient (TP\(^{−/−}\)) mice, the early phase was greatly diminished, whereas the late phase was similar to that apparent in WT mice, suggesting that the TXA\(_2\)-TP system mediates predominantly the early phase of LPS-induced tachycardia (Fig. 4B). In FP-deficient (FP\(^{−/−}\)) mice, LPS induced only the early phase of the increase in heart rate, indicating that the late phase of LPS-induced tachycardia is mediated by the PGF\(_{2α}\)-FP system (Fig. 4B). Finally, the heart rate of LPS-treated TP\(^{−/−}\) FP\(^{−/−}\) mice did not differ substantially from that of vehicle-treated wild-type mice, indicating that TXA\(_2\) and PGE\(_2\) indeed mediate all components of LPS-induced tachycardia in vivo (Fig. 4C). Pretreatment of wild-type mice with indomethacin prevented the effect of LPS on heart rate (Fig. 4C), confirming the role of prostanoids in LPS-
induced tachycardia. Further analyses of the isolated right atrium revealed that both TXA\textsubscript{2} and PGF\textsubscript{2\alpha} directly act on the atrium to increase beating-rate and these actions are independent of adrenergic or muscarinic signaling. Thus the critical roles of prostanoids in tachycardia under systemic inflammatory conditions were demonstrated.

### Arthritis

Rheumatoid arthritis (RA) is a chronic inflammatory disease of the joint characterized by inflammatory cell infiltration, synovial lining hyperplasia, and destruction of cartilage and bone. The importance of PGs in the pathogenesis of RA has long been recognized through the wide use of NSAIDs for RA treatment\textsuperscript{35,36}. Indeed, a large amount of PGI\textsubscript{2} and PGE\textsubscript{2} was detected in the synovial fluid of arthritic joints, suggesting the actions of these PGs in inflammatory sites\textsuperscript{37}. However, how each PG works in RA had remained unknown. Collagen-induced arthritis (CIA) and collagen antibody-induced arthritis (CAIA) are the widely used arthritis models in the mouse\textsuperscript{38}. CIA is induced by immunizing mice with anti-type II collagen (CII) antibody, whereas CAIA is induced by the administration of a combination of monoclonal anti-CII antibodies and LPS. CAIA can be induced in various mouse strains with rapid onset compared with that of CIA. However, the lesions of CAIA are milder and its symptoms last for a shorter duration than CIA\textsuperscript{39}. On the other hand, although the induction of CIA is limited to a few mouse strains such as DBA/1J and takes about a month to develop, its lesions last for a long time and its histopathology, characterized by synovitis, pannus formation, cartilage erosion, and bone destruction in joints, is quite similar to that of human RA. Therefore, CIA is suitable for analyzing chronic joint inflammation. In CAIA model, the importance of the EP\textsubscript{1} was demonstrated, because EP\textsubscript{1} mice showed a profound decrease in the intensity of the inflammation, as well as a decrease in markers of joint destruction, compared to that in the other EP subtype receptor null mice\textsuperscript{40}. In CIA model, COX-2\textsuperscript{-/-} mice display significant reductions in synovial inflammation and joint destruction, whereas arthritis in COX-1\textsuperscript{-/-} mice is indistinguishable from controls\textsuperscript{41}. Moreover mice deficient in microsomal PGE synthase(mPGES-1) showed reduced arthritic responses\textsuperscript{42}, suggesting the importance of PGE\textsubscript{2}-EP\textsubscript{1} signalling in CIA model. However, loss or inhibition of each PGE receptor subtype alone did not affect elicitation of inflammation in CIA\textsuperscript{43}. IP\textsubscript{2} deficient (IP\textsuperscript{2-/-}) mice exhibited significant reduction in arthritic scores compared with WT mice (Fig.5A). Thus, the critical role of PGI\textsubscript{2}-IP signaling in the development of CIA has been clarified. This is an interesting result because PGI\textsubscript{2} has been considered as a mediator of acute inflammation by causing vasodilation or enhancing vascular permeability, and less attention has been paid to the contribution of PGI\textsubscript{2} to chronic inflammation. However, although the CIA score was significantly suppressed by the loss of IP, there was still a small extent of inflammation remaining in IP\textsuperscript{2-/-} mice. To examine if other PG signaling con-
Fig. 5 The roles of IP and EP2/EP4 in collagen-induced arthritis (CIA)

(A) Time course of arthritic scores in WT mice treated with vehicle (closed circle) or in IP− mice treated either with vehicle (open circle) or indomethacin (4 mg/kg/day; open square) in CIA. Vehicle or indomethacin was administered from day 21. * p < 0.05.

(B) Time course of arthritic scores in EP2− mice treated with vehicle (closed circle) or the EP4 antagonist (open circle) in CIA. Vehicle or EP4 antagonist (10 mg/kg/day) was administered from day 21. * p < 0.05 versus vehicle-treated mice.

(C) Effects of the IP agonist (1 μM cicaprost), the EP1 agonist (1 μM ONO-DI-004), the EP2 agonist (1 μM butaprost), the EP3 agonist (1 μM AE248), and the EP4 agonist (1 μM ONO-AE1-329) on IL-6 production in synovial fibroblasts.

(D) Schematic overview of the roles of prostanoid receptor signalling pathways in arthritis. Both the PGD2-IP and PGE2-EP2/EP4 signalling in synovial fibroblasts induces cAMP elevation, which activates these cells to stimulate the induction of various arthritis-related genes, such as IL-6, IL-11, RANKL, VEGF, etc. These mechanisms may work as an amplifier of the inflammatory responses in the arthritic joint.

Fig. 6 The roles of DP and EP3 in ovalbumin (OVA)-induced allergic asthma

(A) Enhanced inflammatory cell infiltration in EP3− mice. Lung histology. The lungs of OVA-sensitized WT or EP3− mice were dissected 24 h after the last inhalation of OVA and were stained with hematoxylin and eosin (left). Scale bars, 50 μm. The area of inflammatory cell infiltration around pulmonary arterioles was quantified in four mice of each group (right). * p < 0.05

(B) A schematic model for functional antagonism between the PGD2-DP pathway and the PGE2-EP3 pathway in development of allergic inflammation associated with asthma. Allergic inflammation consists of two phases, early phase reaction, and the late phase reaction. In the early phase mast cell produce PGD2 upon antigen challenge. PGD2 acts on DP receptor in airway epithelial cells to trigger subsequent allergic reactions. PGE2-EP3 signalling negatively modulates allergic inflammation in both early phase and the late phase reactions (as indicated by −). Thus, the PGD2-DP pathway and the PGE2-EP3 pathway exert opposing actions in allergic inflammation. DP and EP3 are expressed in airway epithelial cells, in which these signals may regulate synthesis and release of chemokines and cytokines.
tributes to this remnant inflammation, IP⁺ mice was treated with indomethacin from day 21. Indomethacin administration abolished the arthritis in IP⁺ mice almost completely, suggesting that PG signaling other than the PGI₂-IP pathway also works in progression of CIA (Fig.5A). Further examination revealed that simultaneous inhibition of EP₂ and EP₃ is needed to achieve a partial but significant suppression of CIA (Fig.5B). These results suggest the hypothesis that EP₂ and EP₃ works redundantly for elicitation of CIA. Consistently, when selective agonist to the IP or each of the EP subtypes were added to the synovial fibroblast culture to examine their activity to enhance the IL-1β-stimulated IL-6 production, the compounds selective to IP, EP₂ and EP₃, but not those to EP₁ and EP₄, potently enhanced the IL-6 production (Fig.5C). Other than IL-6, PGI₂-IP signaling induced a variety of arthritis-related genes, such as IL-11, VEGF, FGF-2, and RANKL, suggesting that the IP-dependent activation of synovial fibroblasts plays a significant role in the effector mechanisms of inflammation in the joint. It should be noted that IP signaling can induce expression of these genes only in combination with IL-1/β, and the stimulation of IP alone induces a significant but marginal effect. Thus, the PGI₂-IP signaling in synovial fibroblasts works as an amplifier of the inflammatory processes in the joint. Because EP₂/EP₃ receptor shares the cAMP signaling pathway with IP, it is quite likely that they also work as an amplifier. Collectively, both PGI₂-IP and PGE₂-EP₂/EP₃ signalling play significant roles in the development of CIA, though it has long been thought that PGE₂ is the primary PG responsible for inflammation in RA. This suggests that inhibition of PGE₂ synthesis alone may not be sufficient for suppression of RA symptoms, providing a cautious note that mPGES inhibitors may not be so effective in RA as COX-2 inhibitors.

**Allergic Asthma**

The roles of prostanoids in allergy had been less well defined than those in acute inflammation, in part because the effects of NSAIDs are far less marked. Nevertheless, allergic responses are associated with an increase in prostanoid formation. For example, PGD₂ is a major prostanoid generated by mast cells upon allergen challenge and is produced abundantly in allergic diseases such as asthma, allergic dermatitis, and conjunctivitis. Little attention had been paid to the roles of PGD₂ in allergy. Matsuoka et al. ⁴⁴ examined this issue by subjecting DP-deficient (DP⁻⁻) mice to ovalbumin (OVA)-induced allergic asthma. They found a marked reduction in the airway inflammation, obstruction, and hypersensitivity in DP⁺⁺ animals, suggesting that PGD₂, acting via the DP, works as a mediator of allergy. On the other hand, Fujitani et al. ⁴⁵ used transgenic (TG) mice overexpressing human lipocalin-type PGD synthase to examine the effect of overproduction of PGD₂ in an asthma model. They showed that these TG mice demonstrated the enhanced accumulation of eosinophils and lymphocytes in the lung accompanied by an increase in the levels of Th2 cytokines and a chemokine, CCL11. In relation with human asthma, Oguma et al. ⁴⁶ demonstrated the significant association between functional genetic variants of the prostanoid DP receptor gene (PTGDR) and susceptibility to asthma. In this study they identified several single-nucleotide polymorphisms (SNPs) in PTGDR and its vicinity. The defined SNPs are associated with the reduced gene expression with reduced transcriptional efficiency of PTGDR and a lower risk of asthma in humans. Recently developed DP receptor antagonist, S-5751, was reported to inhibited the allergic symptoms dramatically in guinea pig models of allergic rhinitis, conjunctivitis and asthma ⁴⁷. These findings provide evidences that PGD₂-DP signalling mediates allergic reactions and that DP receptor antagonists may be useful in the treatment of allergic diseases.

The finding of pro-inflammatory roles of PGD₂-DP signaling raises a new question of why aspirin is not beneficial in allergy and can even precipitate asthmatic attacks in certain individuals. This suggests that other prostanoids normally antagonize the action of PGD₂, making NSAIDs treatment have complex effects on the disease pathway. To address this issue Kunikata et al. ⁴⁸ examined each of EP receptor subtype null mice. EP₂-deficient (EP₂⁻⁻) mice developed more pronounced allergic inflammation than that in WT or other EP subtype receptor null mice (Fig.6A). Conversely, an EP₃-selective agonist suppressed the inflammation. This suppression was effective even when the agonist was administered 3 h after antigen challenge and was associated with inhibition of allergy-related gene expression. Thus, the PGE₂-EP₃ pathway is an important negative modulator of allergic reactions. Since EP₁ is expressed in airway epithelial cells where the chemokines, including CCL11 and CCL17, are also expressed, PGE₂-EP₃ signalling may directly suppress the expression of these chemokine genes. On the basis of these findings, we suggested that PGE₂ produced during allergy acts at EP₁ on both mast cells and airway epithelial cells, thereby blunting activation of mast cells and impeding progression of the allergic reaction by inducing down-regulation of the expression of relevant genes in the airway epithelium (Fig.6B). DP is also expressed in the airway epithelium, and, given its opposing mechanism of signal transduction
relative to that of EP3, it may facilitate the asthmatic reaction by increasing expression of these latter genes. Recently, it was reported that exposure to aerosolized PGD2 before challenge induce the expression of macrophage-derived chemokine (MDC/CCL22), a chemoattractant for Th2 cells, and accelerates Th2 type inflammation\(^{69}\). In view of molecular mechanisms of PGD2-DP signalling in allergic asthma, it is reported that PGD2-DP signalling is mediated via p38 MAPK, p44/42 MAPK, and PKC in a cell type-specific manner leading to NF-κB activation stimulating COX-2 gene expression\(^{69}\).

Conclusion

In this review, we have shown several recent findings of the roles of prostanoid receptor signaling in inflammatory and immune responses. These finding were mainly derived from the analyses of the gene knockout mice in animal model of human disease conditions. Although species differences between human and mouse should be considered cautiously, they not only provide the new insights of detailed mechanisms of previously known prostanoid functions, but also uncovered the new functions masked by their opposing functions such as DP and EP\(_3\) signaling in allergic asthma. These are representative examples of a series of studies on the functions of prostanoid receptor signaling using the gene knockout mice. Various kinds of animal model of human disease conditions have been applied on these mice and these analyses have provided the great opportunities to dissect the precise molecular mechanism for each known prostanoid action and answered to various long-standing questions, as well as to uncover the previously unknown prostanoid functions that had not been predicted from the effects of aspirin-like drugs. These newly discovered functions include actions in the brain, the immune system, and allergy, showing how prostanoids delicately regulate various processes in the body and its responses to environmental stimuli. Indeed, they have sometimes opposing actions through complex interaction mechanisms between different prostanoids, target cells, phase of reaction and so on. The importance of each of these signalling pathways can be altered depending upon the inflammatory stimulus, the type of prostanoids predominantly produced, and the profile of prostanoid receptor expressions. More specific studies will be required to address these issues. Together with recently developed specific pharmacological reagents, tissue-specific inducible gene knockout animals will be extremely powerful tools to dissect the precise mechanisms for elucidating the roles of these signaling pathways in each disease condition. These studies certainly facilitate a search for new applications of the highly selective agonists and antagonists to human disease conditions.

Acknowledgments

This work was supported in part by Grants-in-Aid for Scientific Research from the Ministry of Education Culture Sports Science and Technology of Japan, by a grant from the National Institute of Biomedical Innovation of Japan, and by grants from the Kowa Life Science Foundation, the Takeda Scientific Foundation and the ONO Research Foundation.

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