Chapter 13 Prostaglandins, Leukotrienes (Eicosanoids) and Platelet Activating Factor

PROSTAGLANDINS AND LEUKOTRIENES (Eicosanoids)

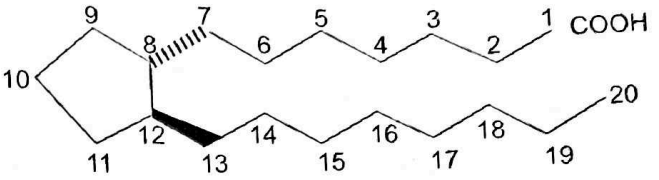
Prostaglandins (PGs) and Leukotrienes (LTs) are biologically active derivatives of 20 carbon atom polyunsaturated essential fatty acids that are released from cell membrane phospholipids. They are the major lipid derived autacoids.

In the 1930s human semen was found to contract isolated uterine and other smooth muscle strips and to cause fall in BP in animals. The active principle was termed 'prostaglandin', thinking that it was derived from prostate. Only in the 1960s it was shown to be a mixture of closely related compounds, the chemical structures were elucidated and widespread distribution in the body was revealed. In 1970s it became clear that aspirin like drugs act by inhibiting PG synthesis, and that in addition to the classical PGs (PGEs and PGFs), thromboxane (TX), prostacyclin (PGI) and leukotrienes (LTs) were of great biological importance. Bergstrom, Samuelsson and Vane got the Nobel prize in 1982 for their work on PGs and LTs. Over the past 40 years PGs and LTs have been among the most intensely investigated substances.

CHEMISTRY, BIOSYNTHESIS AND DEGRADATION

Chemically, PGs may be considered to be derivatives of *prostanoic acid,* though prostanoic acid does not naturally occur in the body. It has a five membered ring and two side chains projecting in opposite directions at right angle to the plane of the ring. There are many series of PGs and thromboxanes (TXs) designated A, B, C....I, depending on the ring structure and the substituents on it. Each series has members with subscript 1, 2, 3 indicating the number of double bonds in the side chains.

Leukotrienes are so named because they were first obtained from leukocytes (*leuko*) and have



PROSTANOIC ACID

3 conjugated double bonds (*triene*). They have also been similarly designated A, B, C.....F and given subscripts 1, 2, 3, 4.

In the body PGs, TXs and LTs are all derived from eicosa (referring to 20 C atoms) tri/tetra/penta enoic acids. Therefore, they can be collectively called *eicosanoids*. In human tissues, the fatty acid released from membrane lipids in largest quantity is *5,8,11,14 eicosa tetraenoic acid (arachidonic acid)*. During PG, TX and prostacyclin synthesis, 2 of the 4 double bonds of arachidonic acid get saturated in the process of cyclization, leaving 2 double bonds in the side chain. Thus, subscript 2 PGs are the most important in man, e.g. PGE_2, $PGF_{2\alpha}$, PGI_2, TXA_2. No cyclization or reduction of double bonds occurs during LT synthesis—the LTs of biological importance are LTB_4, LTC_4, LTD_4.

Eicosanoids are the most universally distributed autacoids in the body. Practically every cell and tissue is capable of synthesizing one or more types of PGs or LTs. As such, they have potent and a very wide range of biological activity. The pathways of biosynthesis of eicosanoids are summarized in Fig. 13.1.

There are no preformed stores of PGs and LTs. They are synthesized locally and the rate of synthesis is governed by the rate of release of arachidonic acid from membrane lipids in

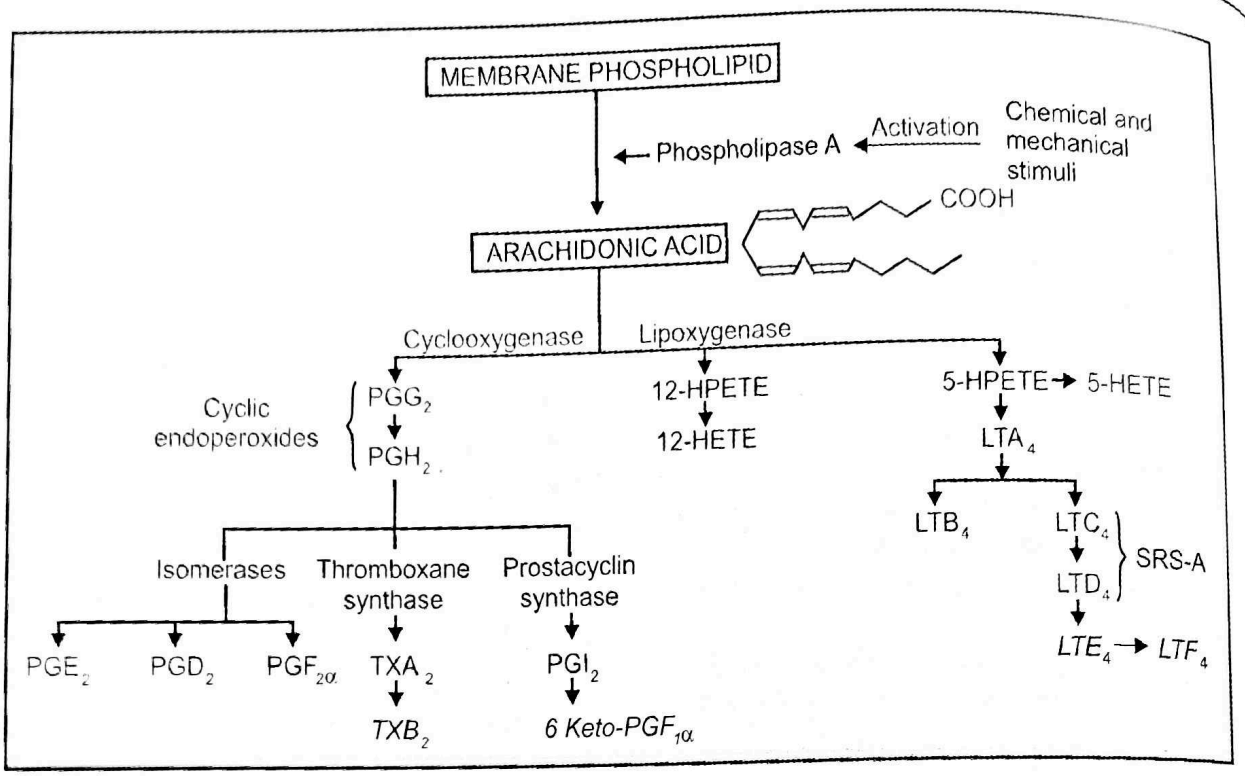


Fig. 13.1: Biosynthesis of prostaglandins (PGs) and leukotrienes (LTs). Less active metabolites are shown in italics TX—Thromboxane; PGI—Prostacyclin; HPETE—Hydroperoxy eicosatetraenoic acid (Hydroperoxy arachidonic acid); HETE—Hydroxyeicosatetraenoic acid (Hydroxy arachidonic acid); SRS-A—Slow reacting substance of anaphylaxis

SECTION 3

response to appropriate stimuli. These stimuli activate hydrolases, including phospholipase A, probably through increased intracellular Ca^{2+}.

Cyclooxygenase (COX) *pathway* It generates eicosanoids with a ring structure (PGs, TXs, prostacyclin) while *lipoxygenase* (LOX) produces open chain compounds (LTs). All tissues have COX—can form cyclic endoperoxides PGG_2 and PGH_2 which are unstable compounds. Further course in a particular tissue depends on the type of isomerases or other enzymes present in it. PGE_2 and $PGF_{2\alpha}$ are the primary prostaglandins (name based on the separation procedure: PGE partitioned into *Ether* while PGF into phosphate [*Fosfat* in Swedish] buffer; α in $PGF_{2\alpha}$ refers to orientation of OH group on the ring). PGs A, B and C are not found in the body: they are artifacts formed during extraction procedures. Lung and spleen can synthesize the whole range of COX products. Platelets primarily synthesize TXA_2 which is—chemically unstable, spontaneously changes to TXB_2. Endothelium mainly generates prostacyclin (PGI_2) which is also chemically unstable and rapidly converts to 6-keto $PGF_{1\alpha}$.

Cyclooxygenase is known to exist in two isoforms COX-1 and COX-2. While both isoforms catalyse the same reactions, COX-1 is a constitutive enzyme in most cells—it is synthesized and is active in the basal state; the level of COX-1 activity is not much changed once the cell is fully grown. On the other hand, COX-2 normally present in insignificant amounts, is inducible by cytokines, growth factors and other stimuli during the inflammatory response. It is believed that eicosanoids produced by COX-1 participate in physiological (house keeping) functions such as secretion of mucus for protection of gastric mucosa, haemostasis and maintenance of renal function, while those produced by COX-2 lead to inflammatory and other pathological changes. However, certain sites in kidney, brain and the foetus constitutively express COX-2 which may play physiological role.

A splice variant of COX-1 (designated COX-3) has been found in the dog brain. This isoenzyme is inhibited by paracetamol and is implicated in the genesis of fever, but the exact role in humans is not known.

Lipoxygenase pathway This pathway appears to operate mainly in the lung, WBC and platelets. Its most important products are the LTs, (generated by 5-LOX) particularly LTB_4 (potent chemotactic) and LTC_4, LTD_4 which together constitute the *'slow reacting substance of anaphylaxis' (SRS-A)* described in 1938 to be released during anaphylaxis. A membrane associated transfer protein called FLAP (five lipoxygenase activating protein) carrys arachidonic acid to 5-LOX, and is essential for the synthesis of LTs. Platelets have only 12-LOX.

Apart from PGs and LTs, a number of other active products can be generated from arachidonic acid at certain sites.

HPETEs produced by LOX can also be converted to *hepoxilins, trioxilins* and lipoxins. A third enzymatic pathway involving cytochrome P450 can metabolize arachidonic acid into *19-* and *20-HETEs* and *epoxyeicosatrienoic acids*. Free radicals can attack arachidonic acid to produce *isoprostanes* nonenzymatically. Brain cells couple arachidonic acid with ethanolamine to produce *anandamide* and a few other related eicosanoids which are now recognized to be the endogenous cannabinoid receptor ligands. Accordingly, they produce cannabis like effects. Like the other eicosanoids, they are synthesized only when needed at the site of action.

Inhibition of synthesis Synthesis of COX products can be inhibited by nonsteroidal anti-inflammatory drugs (NSAIDs). Aspirin acetylates COX at a serine residue and causes irreversible inhibition, while other NSAIDs are competitive and reversible inhibitors. Most NSAIDs are nonselective COX-1 and COX-2 inhibitors, but some later ones like celecoxib, etoricoxib are selective for COX-2.

The sensitivity of COX in different tissues to inhibition by these drugs varies; selective inhibition of formation of certain products may be possible at lower doses. NSAIDs do not inhibit the production of LTs. Rather, LT production may be increased since all the arachidonic acid becomes available to the LOX pathway.

Zileuton inhibits LOX and decreases the production of LTs. It was used briefly in asthma, but has been withdrawn.

Glucocorticosteroids inhibit the release of arachidonic acid from membrane lipids (by enhancing production of proteins called *annexins* which inhibit phospholipase A_2). Production of all eicosanoids—PGs, TXs and LTs is indirectly curtailed. Moreover, steroids inhibit the induction of COX-2 by cytokines at the site of inflammation.

Degradation Biotransformation of arachidonates occurs rapidly in most tissues, but fastest in the lungs. Most PGs, TXA_2 and prostacyclin have plasma t½ of a few seconds to a few minutes. First a specific carrier mediated uptake into cells occurs, the side chains are then oxidized and double bonds are reduced in a stepwise manner to yield inactive metabolites. Metabolites are excreted in urine. PGI_2 is catabolized mainly in the kidney.

ACTIONS AND PATHOPHYSIOLOGICAL ROLES

Prostaglandins, thromboxanes and prostacyclin

The cyclic eicosanoids produce a wide variety of actions depending upon the particular PG (or TX or PGI), species on which tested, tissue, hormonal status and other factors. PGs differ in their potency to produce a given action and different PGs sometimes have opposite effects. Even the same PG may have opposite effects under different circumstances. The important actions of PGs and TXA_2 are summarized in Table 13.1. Since virtually all cells and tissues are capable of forming one or more PGs, these autacoids have been implicated as mediators or modulators of a number of physiological processes and pathological states.

1. CVS PGE_2 causes vasodilatation in most, but not all, vascular beds. $PGF_{2\alpha}$ constricts many larger veins including pulmonary vein and artery. Fall in BP occurs when PGE_2 is injected i.v., but $PGF_{2\alpha}$ has little effect on BP.

- PGI_2 is uniformly vasodilatory and is more potent hypotensive than PGE_2.
- TXA_2 consistently produces vasoconstriction.
- PGE_2 and $F_{2\alpha}$ stimulate heart by weak direct but more prominent reflex action due to fall in BP. The cardiac output increases.

Role

- PGs do not circulate in blood and have no role in regulating systemic vascular resistance. However, PGI_2 generated in the vascular endothelium, mainly by COX-2, appears to be involved in the regulation of local vascular tone as a dilator.
- PGE_2 is continuously produced locally in the ductus arteriosus by COX-2 during foetal life and keeps it patent. At birth its synthesis stops and closure occurs. Aspirin and indomethacin induce closure when it fails to occur spontaneously.
- PGs, generated mainly by COX-2, along with LTs and other autacoids may mediate vasodilatation and exudation at the site of inflammation.

2. Platelets TXA_2, which can be produced locally by platelets, is a potent inducer of aggregation and release reaction. On the other hand PGI_2 (generated by vascular endothelium) is a potent inhibitor of platelet aggregation. PGE_2 has dose dependent pro- and anti-aggregatory effects

Role TXA_2 produced by platelets and PGI_2 produced by vascular endothelium probably constitute a mutually antagonistic system: preventing aggregation of platelets while in circulation and inducing aggregation on injury, when plugging and thrombosis are needed.

Aspirin interferes with haemostasis by inhibiting platelet aggregation. TXA_2 produced by platelet COX-1 plays an important role in amplifying aggregation. Before it is deacetylated in liver, aspirin acetylates COX-1 in platelets while they are in portal circulation. Further, platelets are unable to regenerate fresh COX-1 (lack nucleus: do not synthesize protein), while vessel wall is able to do so (fresh enzyme is synthesized within hours). Thus, at low doses, aspirin selectively inhibits TXA_2 production and has antithrombotic effect lasting > 3 days.

3. Uterus PGE_2 and $PGF_{2\alpha}$ consistently contract human uterus *in vivo*, both pregnant as well as nonpregnant. The sensitivity is higher during pregnancy and there is progressive modest increase with the advance of pregnancy. However, even during early stages, uterus is quite sensitive to PGs though not to oxytocin. PGs increase basal tone as well as amplitude of uterine contractions.

At term, PGs soften the cervix at low doses and make it more compliant.

Role

- Foetal tissues produce PGs. At term $PGF_{2\alpha}$ has been detected in maternal blood. It is postulated that PGs mediate initiation and progression of labour. Aspirin has been found to delay the initiation of labour and also prolong its duration.
- Dysmenorrhoea in many women is associated with increased PG synthesis by the endometrium. This apparently induces uncoordinated uterine contractions which compress blood vessels causing uterine ischaemia and pain. Aspirin group of drugs are highly effective in relieving dysmenorrhoea in most women.

4. Bronchial muscle $PGF_{2\alpha}$, PGD_2 and TXA_2 are potent bronchoconstrictors (more potent than histamine) while PGE_2 is a powerful bronchodilator. PGI_2 produces mild dilatation. Asthmatics are more sensitive to constrictor as well as dilator effects of PGs. PGE_2 and PGI_2 also inhibit histamine release and are effective by aerosol. However, these antiasthmatic effects of PGE_2 and PGI_2 cannot be exploited clinically because they produce irritation of the respiratory tract and have brief action.

Role Asthma may be due to an imbalance between constrictor PGs ($F_{2\alpha}$, PGD_2, TXA_2) and cysteinyl LTs on one hand and dilator ones (PGE_2, PGI_2) on the other. In few individuals aspirin-like drugs consistently induce asthma, possibly by diverting arachidonic acid to produce excess LTC_4 and D_4. This sensitivity

Table 13.1 : A summary of the actions of major prostaglandins, prostacyclin and thromboxane

Organ	*Prostaglandin E_2 (PGE_2)*	*Prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$)*	*Prostacyclin (PGI_2)*	Thromboxane A_2 (TXA_2)
1. *Blood vessels*	Vasodilatation, ↓ BP	Constricts larger veins and some arteries, little effect on BP	Vasodilatation (marked and widespread), ↓↓ BP	Vasoconstriction
2. Heart	Weak inotropic, reflex cardiac stimulation	Weak inotropic	—	—
3. Platelets	Variable effect	—	Antiaggregatory	Aggregation and release reaction
4. Uterus	Contraction (*in vivo*), softening of cervix	Contraction (*in vivo* and *in vitro*), softening of cervix	—	Contraction (*in vitro*)
5. Bronchi	Dilatation, Inhibit histamine release	Constriction	Dilatation (mild), inhibit histamine release	Constriction
6. Stomach	↓ acid secretion, ↑ mucus production	—	↓ acid secretion (weak), mucosal vasodilatation	—
7. Intestine	Contracts longitudinal & relaxes circular muscles, ↑ peristalsis, ↑ Cl^- & water secretion	Spasmogenic, ↑ fluid & electrolyte secretion (weak)	Weak spasmogenic, inhibits toxin-induced fluid secretion	Weak spasmogenic
8. Kidney	Natriuresis, ↓ Cl^- reabsorption, inhibit ADH action, vasodilatation, renin release	—	Natriuresis, vasodilatation, renin release	Vasoconstriction
9. CNS	Pyrogenic, variety of effects on i.c.v. inj.			
10. Afferent nerves	Sensitize to noxious stimuli → tenderness	—	Same as PGE_2	—
11. Endocrine system	Release of ant. pituitary hormones, steroids, insulin; TSH-like action	—	—	
12. Metabolism	Antilipolytic, insulin like action, mobilization of bone Ca^{2+}	—	—	—

is not shared by selective COX-2 inhibitors, indicating that suppression of COX-1 at the pulmonary site is responsible for the reaction. In allergic human asthma, LTs play a more important role, and COX inhibitors are without any effect in most patients.

5. GIT

(i) In isolated preparations, the longitudinal muscle of gut is contracted by PGE_2 and $PGF_{2\alpha}$ while the circular muscle is either contracted (usually by $PGF_{2\alpha}$) or relaxed (usually by PGE_2). Propulsive activity is enhanced in man, especially by PGE_2. This can cause colic and watery diarrhoea, which are important side effects. PGE_2 acts directly on the intestinal mucosa and increases water, electrolyte and mucus secretion.

Role PGs may be involved in mediating toxin induced increased fluid movement in secretory diarrhoeas. In certain diarrhoeas, aspirin can reduce stool volume, but it is not uniformly effective. PGs appear to play a role in the growth of colonic polyps and cancer. Association of lower incidence of colon cancer with regular intake of aspirin is now established. NSAIDs afford relief in familial colonic polyposis by reducing polyp formation.

(ii) PGE_2 markedly reduces acid secretion in the stomach. Volume of juice and pepsin content are also decreased. It inhibits fasting as well as stimulated secretion. Release of gastrin is suppressed (*see* Fig. 47.1). The gastric pH may rise upto 7.0. PGI_2 also reduces gastric secretion, but is less potent. Secretion of mucus and HCO_3^- by gastric mucosal epithelial cells is increased, as is mucosal blood flow. Thus, PGs are antiulcerogenic.

Role PGs (especially PGI_2) appear to be involved in the regulation of gastric mucosal blood flow. They may be functioning as natural ulcer protectives by enhancing gastric mucus and HCO_3^- production, as well as by improving mucosal circulation and health. The ulcerogenic action of NSAIDs appears to be due to loss of this protective influence.

Normally, gastric mucosal PGs are produced by COX-1. Selective COX-2 inhibitors are less ulcerogenic. However, COX-2 gets induced during ulcer healing, and COX-2 inhibitors have the potential to delay healing.

6. Kidney PGE_2 and PGI_2 increase water, Na^+ and K^+ excretion and have a diuretic effect. PGE_2 has a furosemide-like inhibitory effect on Cl^- reabsorption as well. They cause renal vasodilatation and inhibit tubular reabsorption. PGE_2 attenuates ADH action, and this adds to the diuretic effect. In contrast, TXA_2 causes renal vasoconstriction. PGI_2, PGE_2 and PGD_2 evoke release of renin.

Role

- PGE_2 and PGI_2 produced mainly by COX-2 in the kidney appear to function as intrarenal regulators of blood flow as well as tubular reabsorption in kidney. Accordingly, the NSAIDs, including selective COX-2 inhibitors, tend to retain salt and water. The diuretic action of furosemide is blunted by indomethacin—indicating a facilitatory role of PGs by increasing renal blood flow and/or augmenting inhibition of tubular reabsorption.
- Renin release in response to sympathetic stimulation, low salt intake and other influences may be facilitated by PGs.

7. CNS PGs injected i.v. penetrate brain poorly, so that central actions are not prominent. However, injected intracerebroventricularly PGE_2 produces a variety of effects—sedation, rigidity, behavioural changes and marked rise in body temperature. PGI_2 also induces fever, but TXA_2 is not pyrogenic.

Role

- PGE_2 may mediate pyrogen induced fever and malaise. Aspirin and other inhibitors of PG synthesis are antipyretic. Pyrogens, including cytokines, released during bacterial infection trigger synthesis of PGE_2 in the hypothalamus which resets the thermostat to cause fever. COX-2 is the major isoenzyme involved; selective COX-2 inhibitors are equally efficacious antipyretics. A role of COX-3 has also been proposed.

PGs may be functioning as neuromodulators in the brain by regulating neuronal excitability. A role in pain perception, sleep and some other functions has been suggested.

8. Sympathetic nerves Depending on the PG, species and tissue, both inhibition as well as augmentation of NA release from sympathetic nerve endings has been observed. However, PGE_2 mostly inhibits NA release.

Role PGs may modulate sympathetic neurotransmission in the periphery.

9. Peripheral nerves PGs (especially E_2 and I_2) sensitize afferent nerve endings to pain inducing chemical and mechanical stimuli (Fig. 13.2). They irritate mucous membranes and produce long lasting dull pain on intradermal injection.

Role PGs serve as algesic agents during inflammation. They cause tenderness and amplify the action of other algesics. Inhibition of PG synthesis is a major antiinflammatory mechanism. Aspirin injected locally decreases pain produced by injection of bradykinin at the same site. Moreover, PGs released in the dorsal horn of spinal cord by painful stimuli sensitise the neurones to pain perception.

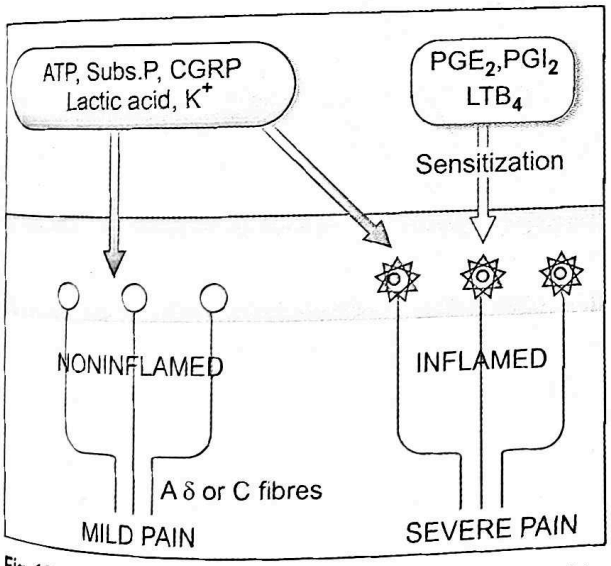


Fig. 13.2: Sensitization of nociceptors (pain receptors) to mediators of pain by prostaglandins at the inflammatory site. Subs. P—Substance P; CGRP—Calcitonin gene related peptide

10. Eye: $PGF_{2\alpha}$ induces ocular inflammation and lowers i.o.t by enhancing uveoscleral and trabecular outflow. Nonirritating congeners like *latanoprost* are now first line drugs in wide angle glaucoma (*see* p. 169).

Role Locally produced PGs appear to facilitate aqueous humor drainage. The finding that COX-2 expression in the ciliary body is deficient in wide angle glaucoma patients supports this contention.

11. Endocrine system PGE_2 facilitates the release of anterior pituitary hormones (growth hormone, prolactin, ACTH, FSH and LH) as well as that of insulin and adrenal steroids. It has a TSH-like effect on the thyroid.

12. Metabolism PGEs are antilipolytic, exert an insulin like effect on carbohydrate metabolism and mobilize Ca^{2+} from bone. They may facilitate hypercalcaemia due to bony metastasis.

Leukotrienes

The straight chain lipoxygenase products of arachidonic acid are produced by a more limited number of tissues (LTB_4 mainly by neutrophils; LTC_4 and LTD_4—the cysteinyl LTs—mainly by macrophages), but probably they are pathophysiologically as important as PGs.

1. CVS and blood LTC_4 and LTD_4 injected i.v. evoke a brief rise in BP followed by a more prolonged fall. The fall in BP is not due to vasodilatation because no relaxant action has been seen on blood vessels. It is probably a result of coronary constriction induced decrease in cardiac output and reduction in circulating volume due to increased capillary permeability. These LTs markedly increase capillary permeability and are more potent than histamine in causing local edema formation.

LTB_4 is highly chemotactic for T-lymphocytes, neutrophils and monocytes; this property is not shared by other LTs. Migration of neutrophils through capillaries and their clumping at sites of inflammation in tissues is also promoted by LTB_4. The cysteinyl LTs (C_4, D_4) are chemotactic for eosinophils and T-lymphocytes.

Role LTs are important mediators of inflammation. They are produced (along with PGs) locally at the site of injury. While LTC_4 and D_4 cause exudation of plasma, LTB_4 attracts the

inflammatory cells which reinforce the reaction. They play greater role in chronic inflammatory states. 5-HPETE and 5-HETE may facilitate local release of histamine from mast cells.

2. Smooth muscle LTC_4 and D_4 contract most smooth muscles. They are potent bronchoconstrictors and induce spastic contraction of g.i.t. at low concentrations.

They also increase mucus secretion in the airways.

Role The cysteinyl LTs (C_4 and D_4) are the most important mediators of human allergic asthma. They are released along with PGs and other autacoids during AG: AB reaction in the lungs. In comparison to other mediators, they are more potent and are metabolized slowly in the lungs, therefore exert a long lasting action. LTs may also be responsible for abdominal colics during systemic anaphylaxis.

3. Afferent nerves Like PGE_2 and I_2, the LTB_4 also sensitizes afferents carrying pain impulses—contributes to pain and tenderness of inflammation.

PROSTANOID RECEPTORS

PGs, TX and prostacyclin act on their own specific receptors located on cell membrane. Five families of prostanoid receptors (DP, EP, FP, IP and TP) have been designated, each after the endogenous PG for which it has the greatest affinity. This has been supported by receptor cloning. All prostanoid receptors are G-protein coupled receptors which can be functionally categorized into 'excitatory' or 'contractile', 'relaxant' and 'inhibitory' groups (Fig. 13.3).

The *contractile group* (EP_1, FP, TP) couple primarily with Gq protein and activate PLC_β

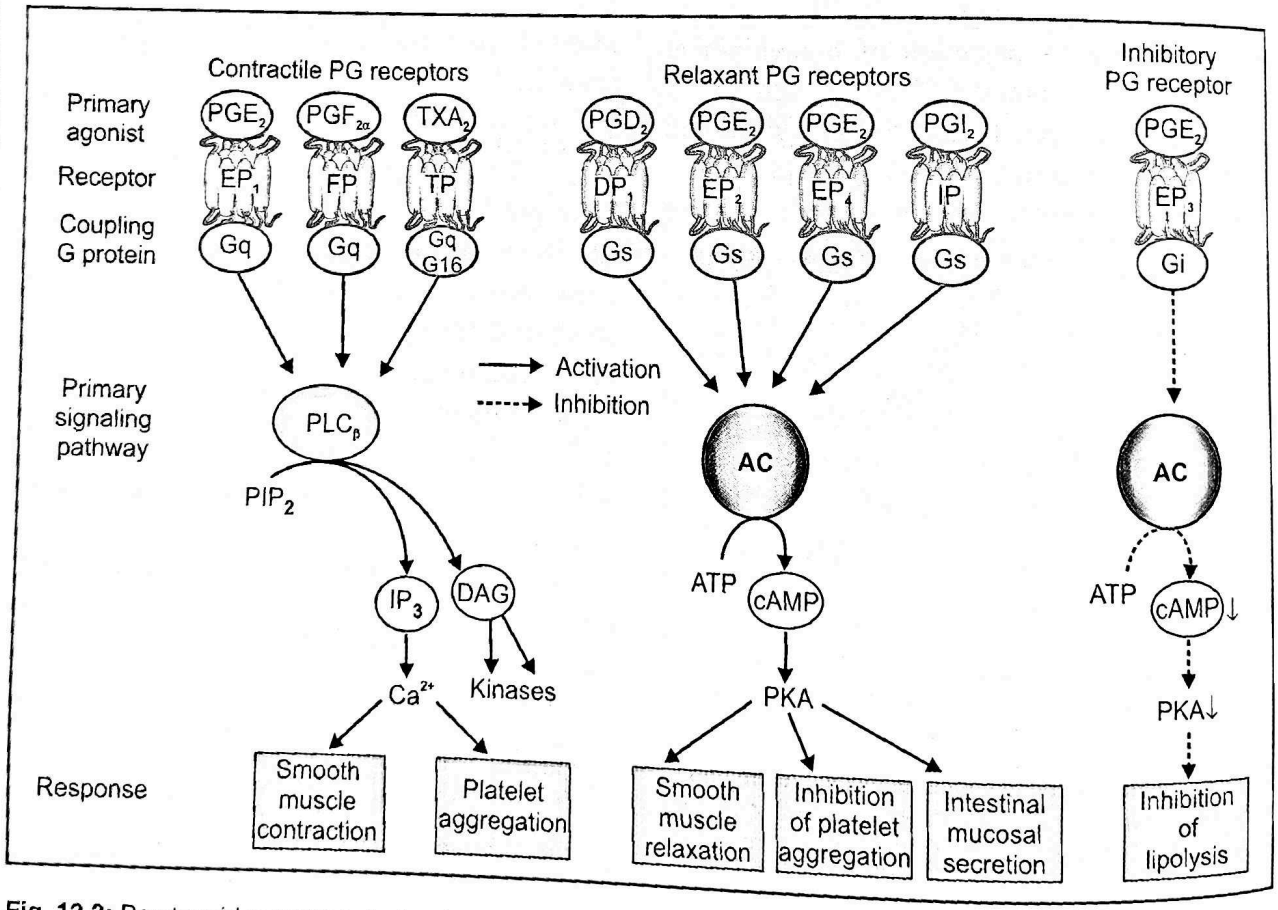


Fig. 13.3: Prostanoid receptors, their primary signaling pathways and major responses elicited through them. All prostanoid receptors are G-protein coupled receptors. On the basis of their functional characterization, prostanoid receptors have been grouped into *contractile, relaxant* and *inhibitory* groups.
PLC_β—Phospholipase Cβ; IP_3—Inositol trisphosphate; DAG—Diacyl glycerol; AC—Adenylyl cyclase; cAMP—Cyclic AMP; PKA—Protein kinase A.

to generate IP_3 and DAG. These second messengers release Ca^{2+} intracellularly resulting in excitatory responses like smooth muscle contraction, platelet aggregation, etc. The *relaxant group* (DP_1, EP_2, EP_4 and IP) couple with Gs protein—activate adenylyl cyclase to generate intracellular second messenger cAMP. Smooth muscle relaxation, inhibition of platelet aggregation, etc. are produced through cAMP dependent protein kinase (PK_A). The *inhibitory* (EP_3) receptor couples with Gi protein, inhibits adenylyl cyclase and exerts antilipolytic action. The major characteristics of subtypes of prostanoid receptors are:

DP This receptor has strongest affinity for PGD_2, but PGE_2 can also activate it. Two subtypes DP_1 and DP_2 have been identified, but both have limited distribution in the body; DP_1 is a relaxant receptor which dilates certain blood vessels and inhibits platelet aggregation. The DP_2 receptor couples with Gi protein and inhibits cAMP generation.

EP This receptor is characterized by highest affinity for PGE_2; *enprostil* is a selective agonist. Four subtypes have been recognized:

EP_1 is a contactile receptor—contracts visceral smooth muscle, but is less abundant in the body.

EP_2 *and* EP_4 are relaxant in nature, act by increasing cAMP in smooth muscle, but the same second messenger enhances Cl^- and water secretion by the intestinal mucosa. While EP_2 is present in few organs, EP_4 has wide distribution.

EP_3 is inhibitory, decreases cAMP generation by coupling with Gi protein. The antilipolytic action of PGE_2 is exerted by opposing cAMP generation in adipose tissue. Distribution of EP_3 receptor in the body is wide.

FP This contactile receptor is highly expressed in the female genital tract, and is present in many other organs. It exhibits strong affinity for $PGF_2\alpha$; *fluprostenol* is a selective agonist.

IP This relaxant receptor is defined by highest affinity for PGI_2, but PGE_2 also acts on it; *cicaprost* is a selective agonist. It is expressed in heart, lungs, kidney, platelet (antiaggregatory), etc., but the highest density is in the vasculature.

TP Characterized by high affinity for TxA_2, this contractile receptor is abundant in platelets (aggregatory), cardiovascular system, immune cells and many other organs. PGH_2 can also activate TP. The TP receptor functions by activating PLC_β through G_q and G_{16} proteins. Apart from IP_3/DAG—Ca^{2+}—PKc pathway, it utilizes other kinases as well to exert certain biological effects.

LEUKOTRIENE RECEPTORS

Separate receptors for LTB_4 (BLT_1 and BLT_2) and for the cysteinyl LTs (LTC_4, LTD_4) have been defined. Two subtypes, $cysLT_1$ and $cysLT_2$ of the cysteinyl LT receptor have been cloned. All LT receptors couple with Gq protein and function through the IP_3/DAG transducer mechanism. The BLT receptors are chemotactic and primarily expressed in leucocytes and spleen. BLT_1 receptor has high, while BLT_2 receptor has lower affinity for LTB_4. The $cysLT_1$ receptor is mainly expressed in bronchial and intestinal muscle and has higher affinity for LTD_4 than for LTC_4. The primary location of $cysLT_2$ receptor is leucocytes and spleen, and it shows no preference for LTD_4 over LTC_4. The $cysLT_1$ receptor antagonists, *viz. Montelukast, Zafirlukast,* etc. are now valuable drugs for bronchial asthma (*see* Ch. 16).

USES

The clinically useful natural and synthetic prostaglandins are listed in the chart below.

PROSTAGLANDINS (PGs)

Natural PGs	Prostaglandin analogues
Dinoprostone (PGE_2)	Carboprost (15-methyl $PGF_{2\alpha}$)
Gemeprost	Misoprostol (methyl PGE_1 ester)
Dinoprost ($PGF_{2\alpha}$)	Latanoprost (PGE_2 analogue)
Alprostadil (PGE_1)	Travoprost
Prostacyclin (PGI_2) (Epoprostenol)	Bimatoprost

Clinical application of PGs and their analogues is rather restricted because of limited availability, short lasting action, cost and frequent side effects. However, their use in glaucoma and in obstetrics is now commonplace. Their indications are:

1. *Abortion* During the first trimester, termination of pregnancy by transcervical suction is the procedure of choice. Intravaginal PGE_2 pessary inserted 3 hours before attempting dilatation can minimise trauma to the cervix by reducing resistance to dilatation.

Medical termination of pregnancy of upto 7 weeks has been achieved with high success rate by administering mifepristone (antiprogestin) 600 mg orally 2 days before a single oral dose of misoprostol 400 µg. This is now a valuable alternative to suction-evacuation. Uterine contractions are provoked and the conceptus is expelled within the next few hours. Intravaginal misoprostol is now favoured by many in place of oral, because it produces fewer side effects. Sublingual route is also advocated by some experts. Ectopic pregnancy should be ruled out beforehand and complete expulsion should be confirmed afterwards. Uterine cramps, vaginal bleeding, nausea, vomiting and diarrhoea are the common side effects. Methotrexate administered along with misoprostol is also highly successful for inducing abortion in the first few weeks of pregnancy.

PGs have a place in midterm abortion, missed abortion and molar gestation, though delayed and erratic action and incomplete abortion are a problem. The initial enthusiasm has given way to more considered use. PGs convert the oxytocin resistant midterm uterus to oxytocin responsive one: a single extraamniotic injection (PGE_2) followed by i.v. infusion of oxytocin, or intraamniotic ($PGF_{2\alpha}$) with hypertonic solution produces 2nd trimester abortion in a high percentage without undue side effects. Pretreatment with mifepristone improves the efficacy of PGE_2 as abortifacient.

2. *Induction/augmentation of labour* PGs do not offer any advantage over oxytocin for induction of labour at term. They are less reliable and show wider individual variation in action. PGE_2 and $PGF_{2\alpha}$ (rarely) have been used in place of oxytocin in toxaemic and renal failure patients, because PGs do not cause fluid retention that is possible with oxytocin. PGE_2 may also be used to augment labour if it is slow, particularly in primipara. Intravaginal route is preferred now because side effects are milder; extra/intra amniotic route is infrequently used.

3. *Cervical priming (ripening)* Applied intravaginally or in the cervical canal, low doses of PGE_2, which do not affect uterine motility, make the cervix soft and compliant. This procedure has yielded good results in cases with unfavourable cervix. If needed labour may be induced 12 hours later with oxytocin: chances of failure are reduced.

4. *Postpartum haemorrhage (PPH)* Carboprost (15-methyl $PGF_{2\alpha}$) injected i.m. is an alternative drug for control of PPH due to uterine atony, especially in patients unresponsive to ergometrine and oxytocin.

PGE_2 (Dinoprostone) PROSTIN-E_2 for induction/augmentation of labour, midterm abortion.

Vaginal gel (1 mg or 2 mg in 2.5 ml) 1 mg inserted into posterior fornix, followed by 1–2 mg after 6 hour if required.

Vaginal tab (3 mg) 3 mg inserted into posterior fornix, followed by another 3 mg if labour does not start within 6 hour.

Extraamniotic solution (10 mg/ml in 0.5 ml amp.) infrequently used.

Intravenous solution (1 mg/ml in 0.75 ml amp., 10 mg/ml in 0.5 ml amp) i.v. route is rarely used due to more side effects.

Oral tablet PRIMIPROST 0.5 mg tab, one tab. hourly till induction, max 1.5 mg per hr; rarely used.

Cervical gel CERVIPRIME (0.5 mg in 2.5 ml prefilled syringe) 0.5 mg inserted into cervical canal for preinduction cervical softening and dilatation in patients with poor Bishop's score.

Gemeprost CERVAGEM 1 mg vaginal pessary: for softening of cervix in first trimester—1 mg 3 hr before attempting dilatation; for 2nd trimester abortion/molar gestation—1 mg every 3 hours, max. 5 doses.

$PGF_{2\alpha}$ (Dinoprost) PROSTIN F_2 ALPHA intraamniotic injection 5 mg/ml in 4 ml amp. for midterm abortion/induction of labour (rarely used).

15-methyl $PGF_{2\alpha}$ (Carboprost) PROSTODIN 0.25 mg in 1 ml amp; 0.25 mg i.m. every 30–120 min for PPH, midterm abortion, missed abortion.

T-PILL + MISO, ABORTOM: Mifepristone 200 mg tab (3 tabs) + misoprostol 200 µg (2 tabs) kit: mifepristone 3 tab orally followed 2 days later by misoprostol 2 tab orally, for termination of pregnancy of upto 49 days.

5. *Glaucoma* Topical $PGF_{2\alpha}$ analogues like *latanoprost, travoprost, bimatoprost* that are FP receptor agonists are the first choice drugs in wide angle glaucoma (*see* p. 169).

6. *Peptic ulcer* Stable analogue of PGE_1 (misoprostol) is occasionally used for healing peptic ulcer, especially

in patients who need continued NSAID therapy or who continue to smoke (*see* Ch. 47).

7. *To maintain patency of ductus arteriosus* in neonates with congenital heart defects, till corrective surgery is undertaken. PGE_1 (Alprostadil) is used; apnoea occurs in few cases.

PROSTIN VR, BIOGLANDIN 0.5 mg in 1 ml amp; dilute and infuse i.v.

8. *To avoid platelet damage* PGI_2 (Epoprostenol) can be used to prevent platelet aggregation and damage during haemodialysis or cardiopulmonary bypass. It also improves harvest of platelets for transfusion.

9. *Pulmonary hypertension* PGI_2 lowers pulmonary artery resistance. Accordingly, few cases of primary *pulmonary hypertension* have been successfully maintained on epoprostenol infused continuously in a large vein.

FLOLAN: epoprostenol 0.5 mg vial for reconstitution.

10. *Peripheral vascular diseases* PGI_2 (or PGE_1) infused i.v. can relieve rest pain and promote healing of ischaemic ulcers in severe cases of intermittent claudication and in Raynaud's disease.

11. *Impotence* Alprostadil (PGE_1) injected into the penis causes erection lasting 1–2 hours. However, oral sildenafil/tadalafil is now preferred for erectile dysfunction.

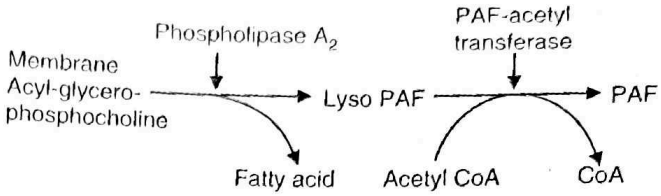
SIDE EFFECTS

Side effects are common in the use of PGs, but their intensity varies with the PG, the dose and the route. These are: nausea, vomiting, watery diarrhoea, uterine cramps, unduly forceful uterine contractions, vaginal bleeding, flushing, shivering, fever, malaise, fall in BP, tachycardia, chest pain.

PLATELET ACTIVATING FACTOR (PAF)

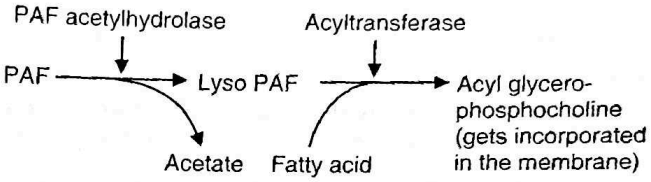
Like eicosanoids, platelet activating factor (PAF) is a cell membrane derived polar lipid with intense biological activity. Discovered in 1970s PAF is active at subnanomolar concentration and is now recognized to be an important signal molecule. PAF is acetyl-glyceryl ether-phosphoryl choline. The ether-linked alkyl chain in human PAF is mostly 16 or 18 C long.

Synthesis and degradation PAF is synthesized from precursor phospholipids present in cell membrane by the following reactions:



The second step is rate limiting. Antigen-antibody reaction and a variety of mediators stimulate PAF synthesis in a Ca^{2+} dependent manner on demand. There are no preformed stores of PAF. In contrast to eicosanoids, the types of cells which synthesize PAF is quite limited—mainly WBC, platelets, vascular endothelium and kidney cells.

PAF is degraded in the following manner:



Actions PAF has potent actions on many tissues/organs.

Platelets PAF induces aggregation and release reaction; also releases TXA_2; i.v. injection of PAF results in intravascular thrombosis.

WBC PAF is a potent chemotactic for neutrophils, eosinophils and monocytes. It stimulates neutrophils to aggregate, to stick to vascular endothelium and migrate across it to the site of infection. It also prompts release of lysosomal enzymes and LTs as well as generation of superoxide radical by the polymorphs. The chemotactic action may be mediated through release of LTB_4. It induces degranulation of eosinophils.

Blood vessels Vasodilatation mediated by release of EDRF occurs causing fall in BP on i.v. injection. Decreased coronary blood flow has been observed on intracoronary injection, probably due to formation of platelet aggregates and release of TXA_2.

PAF is the most potent agent known to increase vascular permeability. Wheal and flare occur at the site of intradermal injection.

Injected into the renal artery PAF reduces renal blood flow and Na^+ excretion by direct vasoconstrictor action, but this is partly counteracted by local PG release.

Visceral smooth muscle Contraction occurs by direct action as well as through release of LTC_4, TXA_2 and PGs. Aerosolized PAF is a potent bronchoconstrictor. In addition, it produces mucosal edema, secretion and a delayed and long-lasting bronchial hyper-responsiveness. It also stimulates intestinal and uterine smooth muscle.

Stomach PAF is highly ulcerogenic: erosions and mucosal bleeding occur shortly after i.v. injection of PAF. The gastric smooth muscle contracts.

Mechanism of action Membrane bound specific PAF receptors have been identified. The PAF receptor is a

G-protein coupled receptor which exerts most of the actions by coupling with Gq protein and generating intracellular messengers $IP_3/DAG \rightarrow Ca^{2+}$ release. It can also inhibit adenylyl cyclase by coupling with Gi protein.

As mentioned above, many actions of PAF are mediated/augmented by PGs, TXA_2 and LTs which may be considered its extracellular messengers. PAF also acts intracellularly, especially in the endothelial cells. Rise in PAF concentration within the endothelial cells is associated with exposure of neutrophil binding sites on their surface. Similarly, its proaggregatory action involves unmasking of fibrinogen binding sites on the surface of platelets.

PAF antagonists A number of natural and synthetic PAF receptor antagonists have been investigated. Important among these are ginkgolide B (from a Chinese plant), and some structural analogues of PAF. The PAF antagonists have manyfold therapeutic potentials like treatment of stroke, intermittent claudication, sepsis, myocardial infarction, shock, g.i. ulceration, asthma and as contraceptive. Some of them have been tried clinically but none has been found worth marketing. Alprazolam and triazolam antagonize some actions of PAF.

Pathophysiological roles PAF has been implicated in many pathological states and some physiological processes by mediating cell-to-cell interaction. These are:

1. *Inflammation:* Generated by leukocytes at the site of inflammation PAF appears to participate in the causation of vasodilatation, exudation, cellular infiltration and hyperalgesia.
2. *Bronchial asthma:* Along with LTC_4 and LTD_4, PAF appears to play a major role by causing bronchoconstriction, mucosal edema, recruiting eosinophils and provoking secretions. It is unique in producing prolonged airway hyper-reactivity, so typical of human bronchial asthma.
3. *Anaphylactic (and other) shock conditions:* are associated with high circulating PAF levels.
4. *Haemostasis and thrombosis:* PAF may participate by promoting platelet aggregation.
5. PAF may also play a role in implantation of fertilized ovum, ischaemic states of brain, heart and g.i.t., including g.i. ulceration.

SECTION 3

PROBLEM DIRECTED STUDY

13.1 A full term primigravida presented with labour pains. On examination the BP was 110/70 mm Hg and she was not anaemic. The presentation was vertex, head was engaged, foetal heart sound was normal, there was no cephalopelvic disproportion, no placenta previa, membranes were intact and uterine contractions were adequate. The labour was allowed to progress under observation. After 8 hours the cervix was still firm and not adequately dilated.
(a) Can some medicaion be used to soften the cervix, help its ripening and facilitate delivery? If so, which drug and route of administration should be used?
(b) Given the above findings, is there any contraindication to the use of such medication?
(see Appendix-1 for solution)