Signal transduction is initiated through the action of a Src family–related tyrosine kinase termed Lyn that is constitutively associated with the β chain. Lyn transphosphorylates the canonical immunoreceptor tyrosine-based activation motifs (ITAMs) of the β and γ chains of the receptor, resulting in recruitment of more active Lyn to the β chain and of Syk tyrosine kinase. The phosphorylated tyrosines in the ITAMs function as binding sites for the tandem *src* homology two (SH2) domains within Syk. Syk activates not only phospholipase Cγ, which associates with the linker of activated T cells at the plasma membrane, but also phosphatidylinositol 3-kinase to provide phosphatidylinositol-3,4,5-trisphosphate, which allows membrane targeting of the Tec family kinase Btk and its activation by Lyn. In addition, the Src family tyrosine kinase Fyn becomes activated after aggregation of IgE receptors and phosphorylates the adapter protein Gab2 that enhances activation of phosphatidylinositol 3-kinase. Indeed, this additional input is essential for mast cell activation, but it can be partially inhibited by Lyn, indicating that the extent of mast cell activation is in part regulated by the interplay between these Src family kinases. Activated phospholipase Cγ cleaves phospholipid membrane substrates to provide inositol-1,4,5-trisphosphate (IP_3) and 1,2-diacylglycerols (1,2-DAGs) so as to mobilize intracellular calcium and activate protein kinase C, respectively. The subsequent opening of calcium-regulated activated channels provides the sustained elevations of intracellular calcium required to recruit the mitogen-activated protein kinases, ERK, JNK, and p38 (serine/threonine kinases), which provide cascades to augment arachidonic acid release and to mediate nuclear translocation of transcription factors for various cytokines. The calcium ion–dependent activation of phospholipases cleaves membrane phospholipids to generate lysophospholipids, which, like 1,2-DAG, may facilitate the fusion of the secretory granule perigranular membrane with the cell membrane, a step that releases the membrane-free granules containing the preformed mediators of mast cell effects.

The secretory granule of the human mast cell has a crystalline structure, unlike mast cells of lower species. IgE-dependent cell activation results in solubilization and swelling of the granule contents within the first minute of receptor perturbation; this reaction is followed by the ordering of intermediate filaments about the swollen granule, movement of the granule toward the cell surface, and fusion of the perigranular membrane with that of other granules and with the plasmalemma to form extracellular channels for mediator release while maintaining cell viability.

In addition to exocytosis, aggregation of FcεRI initiates two other pathways for generation of bioactive products, namely, lipid mediators and cytokines. The biochemical steps involved in expression of such cytokines as tumor necrosis factor α (TNF-α), interleukin (IL) 1, IL-6, IL-4, IL-5, IL-13, granulocyte-macrophage colony-stimulating factor (GM-CSF), and others, including an array of chemokines, have not been specifically defined for mast cells. Inhibition studies of cytokine production (IL-1β, TNF-α, and IL-6) in mouse mast cells with cyclosporine or FK506 reveal binding to the ligand-specific immunophilin and attenuation of the calcium ion- and calmodulin-dependent serine/threonine phosphatase, calcineurin.

Lipid mediator generation **(Fig. 376-1)** involves translocation of calcium ion–dependent cytosolic phospholipase A_2 to the outer nuclear membrane, with subsequent release of arachidonic acid for metabolic processing by the distinct prostanoid and leukotriene pathways. The constitutive prostaglandin endoperoxide synthase-1 (PGHS-1/cyclooxygenase-1) and the de novo inducible PGHS-2 (cyclooxygenase-2) convert released arachidonic acid to the sequential intermediates, prostaglandins G_2 and H_2. The glutathione-dependent hematopoietic prostaglandin D_2 (PGD_2) synthase then converts PGH_2 to PGD_2, the predominant mast cell prostanoid. The PGD_2 receptor DP_1 is expressed by platelets and epithelial cells, whereas DP_2 is expressed by T_H2 lymphocytes, eosinophils, and basophils. Mast cells also generate thromboxane A_2 (TXA_2), a short lived but powerful mediator that induces bronchoconstriction and platelet activation through the T prostanoid (TP) receptor.

For leukotriene biosynthesis, the released arachidonic acid is metabolized by 5-lipoxygenase (5-LO) in the presence of an integral nuclear membrane protein, 5-LO activating protein (FLAP). The calcium ion–dependent translocation of 5-LO to the nuclear membrane converts the arachidonic acid to the sequential intermediates, 5-hydroperoxyeicosatetraenoic acid (5-HPETE) and leukotriene (LT) A_4. LTA_4 is conjugated with reduced glutathione by LTC_4 synthase, an integral nuclear membrane protein homologous to FLAP. Intracellular LTC_4 is released by a carrier-specific export step for extracellular metabolism to the additional cysteinyl leukotrienes, LTD_4 and LTE_4, by the sequential removal of glutamic acid and glycine. Alternatively, cytosolic LTA_4 hydrolase converts some LTA_4 to the dihydroxy leukotriene LTB_4, which also undergoes specific export. Two receptors for LTB_4, BLT_1 and BLT_2, mediate chemotaxis of human neutrophils. Two receptors for the cysteinyl leukotrienes, $CysLT_1$ and $CysLT_2$, are present on smooth muscle of the airways and the microvasculature and on hematopoietic cells such as macrophages, eosinophils, and mast cells. Whereas the $CysLT_1$ receptor has a preference for LTD_4 and is blocked by the receptor antagonists in clinical use, the $CysLT_2$ receptor is equally responsive to LTD_4 and LTC_4, is unaffected by these antagonists, and is a negative regulator of the function of the $CysLT_1$ receptor. LTD_4, acting at $CysLT_1$ receptors, is the most potent known bronchoconstrictor, whereas LTE_4 induces a vascular leak and mediates the recruitment of eosinophils to the bronchial mucosa. Studies in gene-deleted mice indicate the existence of additional receptors for LTE_4. The lysophospholipid formed during the release of arachidonic acid from 1-*O*-alkyl-2-acyl-*sn*-glyceryl-3-phosphorylcholine can be acetylated in the second position to form platelet-activating factor (PAF). Serum levels of PAF correlated positively with the severity of anaphylaxis to peanut in a recent study, whereas the levels of PAF acetyl hydrolase (a PAF-degrading enzyme) were inversely related to the same outcome.

Unlike most other cells of bone marrow origin, mast cells circulate as committed progenitors lacking their characteristic secretory granules. These committed progenitors express c-*kit*, the receptor for stem cell factor (SCF). Unlike most other lineages, they retain and

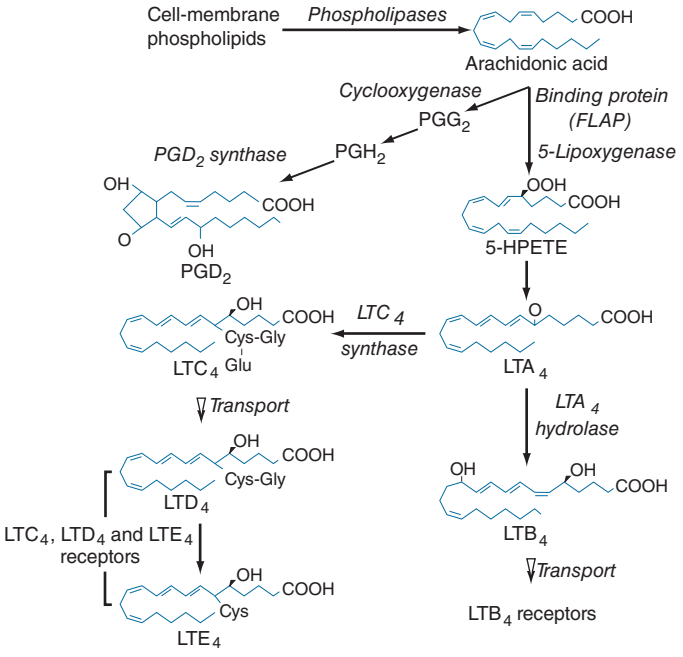

FIGURE 376-1 Pathways for biosynthesis and release of membrane-derived lipid mediators from mast cells. In the 5-lipoxygenase pathway, leukotriene A_4 (LTA_4) is the intermediate from which the terminal-pathway enzymes generate the distinct final products, leukotriene C_4 (LTC_4) and leukotriene B_4 (LTB_4), which leave the cell by separate saturable transport systems. Gamma glutamyl transpeptidase and a dipeptidase then cleave glutamic acid and glycine from LTC_4 to form LTD_4 and LTE_4, respectively. The major mast cell product of the cyclooxygenase system is PGD_2.