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PROSTAGLANDINS AS MEDIATORS OF COX-2 DERIVED CARCINOGENESIS IN GASTROINTESTINAL TRACT

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This review was designed to show the role of expression of cyclooxygenase (COX)-1 and COX-2 in the cancerogenesis of esophagus, stomach and colon. Unlike COX-1, which is expressed in the normal esophageo-gastro-colonic mucosa, COX-2 was found to be expressed mainly in the pre-cancer changes in the mucosa including Barrett's esophagus, *Helicobacter pylori* (*H. pylori*)-induced gastritis and inflammatory changes in colonic mucosa. In Barrett's esophagus, prostaglandins (PGs) derived from upregulated COX-2 contribute to the progression of low-grade to high-grade dysplasia and finally to cancer. In chronic gastritis induced by chronic *H. pylori* infection, overexpression of COX-2 is probably induced by inflammatory cytokines, growth factors, especially gastrin and reactive oxygen species leading to mutagenesis and subsequent metaplasia, dysplasia and cancer formation. The imbalance between cell proliferation and apoptosis caused mainly by products of COX-2 leads to cancerogenesis. Similarly, in colorectal cancer the overexpression of COX-2, possibly induced by the action of growth promoting factors including progastrin and gastrin and overexpression of survivin contribute to the colorectal cancerogenesis that could be, at least in part, amended by the treatment with specific COX-2 inhibitors. We conclude that: 1) COX-2-derived PGs play a key role in the tumorogenesis in the gastrointestinal tract; 2) The tumor-promoting effect of PGs may be attributed to their ability to stimulate cell proliferation and migration, to inhibit the apoptosis and to increase angiogenesis and invasiveness; 3) In accordance to the proposed major role of COX-2 in cancerogenesis, selective COX-2 inhibitors have been shown in numerous studies to exhibit strong chemopreventive effect on the development of gastrointestinal cancers.

Key words: *COX-1, COX-2, prostaglandins, gastrointestinal cancerogenesis, apoptosis*

Prostaglandins (PGs) are hormone-like bioactive substances mediating autocrine and paracrine signaling over short distances and are involved in many physiological and pathological processes. PGs are derived mainly from arachidonic acid released from membrane by phospholipases, mainly phospholipase A2 (PLA2). After liberation from phospholipids, free intracellular arachidonic acid can be further metabolized by three different pathways: PGG/H synthase (better known as cyclooxygenase), lipoxygenase and the cytochrome p-450 monooxygenase (1).

Cyclooxygenase (COX) is a rate-limiting enzyme in the synthesis of PGs. It catalyses the conversion of arachidonic acid to PGG₂, then to PGH₂ which is subsequently converted to various physiologically active prostanoids, including PGE₂, PGD₂, PGF₂α, PGI₂ (prostacyclin) and thromboxane A2 (TXA₂) by the relevant enzymes in a variety of cell types. At least 2 isoforms of COX have been identified so far. COX-1 is expressed constitutively in many tissues and PGs produced by COX-1 mediate the "housekeeping" functions such as cytoprotection of gastric mucosa, regulation of renal blood flow and platelet aggregation. In contrast, COX-2 is not detected in most normal tissues, but its expression is rapidly induced by both inflammatory and mitogenic stimuli resulting in increased synthesis of PGs in inflamed and neoplastic tissue (2) (*Fig. 1*). Recently, a novel COX-1 splice variant termed COX-3 (or recently COX-1b) has been identified in canine tissue (most abundant in cerebral cortex) as an

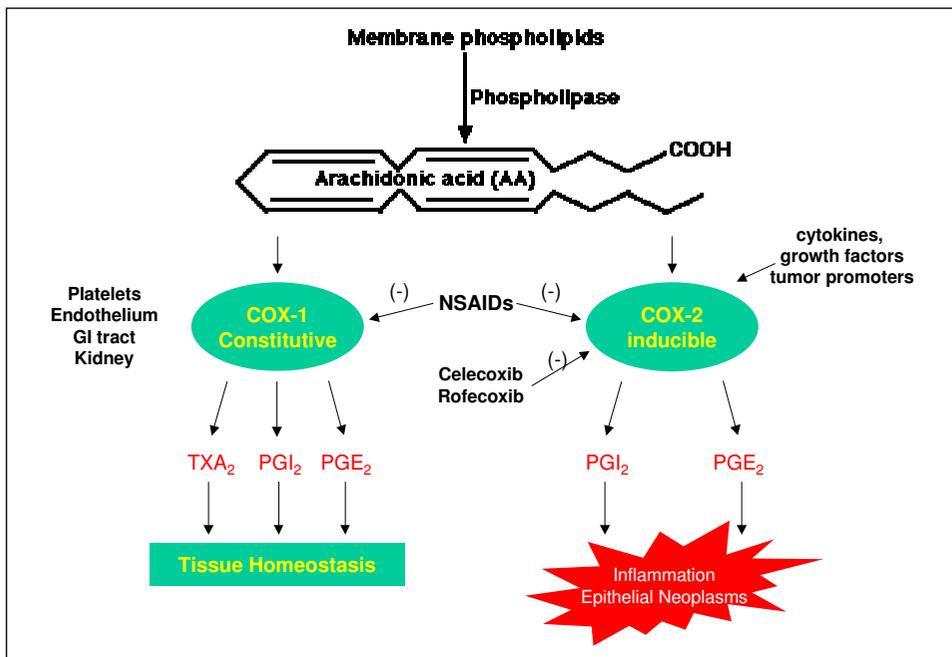


Fig. 1. Schematic presentation of the action of cyclooxygenases (COX-1 and COX-2).

acetaminophen-sensitive isoform. However, the implication of this splice variant in humans is still not known (3).

Enzymatic conversion of arachidonic acid is a major pathway for the synthesis of conventional prostaglandins. In addition, cyclopentenone PGs such as PGA_2 , PGA_1 and PGJ_2 are formed by dehydration within the cyclopentane ring of PGE_2 , PGE_{2v} and PGD_2 , respectively. PGJ_2 can isomerize to yield 9-deoxy- Δ^{12} - PGD_2 (Δ^{12} - PGJ_2). Further dehydration of this molecule results in the formation of 15-deoxy- $\Delta^{12,14}$ - PGJ_2 (15d- PGJ_2). 15d- PGJ_2 differs from other prostaglandins in several aspects. In particular, 15d- PGJ_2 is recognized as the endogenous ligand for the intranuclear receptor $\text{PPAR}\gamma$ (4).

PGs act *via* high-affinity G-protein-coupled receptors, four EP receptors for PGE_2 termed EP1-EP4, IP receptor for prostacyclin, DP receptor for PGD_2 , FP receptor for $\text{PGF}_2\alpha$. These receptors are linked to different signal transduction pathways (5). In addition, peroxisome proliferator-activated receptors (PPAR) have been identified as novel intracellular PG receptors (6).

The possible involvement of PGs in the carcinogenesis is supported by the presence of increased expression of COX-2 in both premalignant and malignant tissues. There is also evidence that growth factors, oncogenes, cytokines and tumor promoters stimulate COX-2 transcription *via* protein kinase C (PKC) and Ras-mediated signaling (7). Another evidence for the role of PGs in the carcinogenesis emerges from the numerous epidemiological studies which demonstrated that chronic intake of non-steroidal drugs (NSAIDs), especially aspirin, prevents cancer development (8-10).

PGs affect numerous mechanisms that have been implicated in carcinogenesis such as cell proliferation, angiogenesis, apoptosis and mutagenesis (*Fig. 2*).

Prostaglandins and cell proliferation

Previous studies including our own have demonstrated that PGs stimulate proliferation of different cell lines derived from gastrointestinal tract such as colonic, intestinal, gastric and esophageal cell lines. Therefore, it is not surprising that NSAIDs as inhibitors of PG synthesis exert inhibitory effect on proliferation of malignant cell lines derived from gastrointestinal tract (*in vitro* studies) and on tumor growth *in vivo* (11). Interestingly, many non-COX-pathways have been identified *in vitro*, and the growth of cells lacking COX expression can also be inhibited by these agents. This means that the anticancer activity of NSAIDs is partly independent of their COX-2 inhibitory properties. The mechanisms by which NSAID inhibit cell proliferation include cell cycle arrest and induction of apoptosis and necrosis (12).

Prostaglandins and apoptosis

Apoptosis, the morphologically defined form of programmed cell death, plays a crucial role in the carcinogenesis. The deregulation of this process can lead to

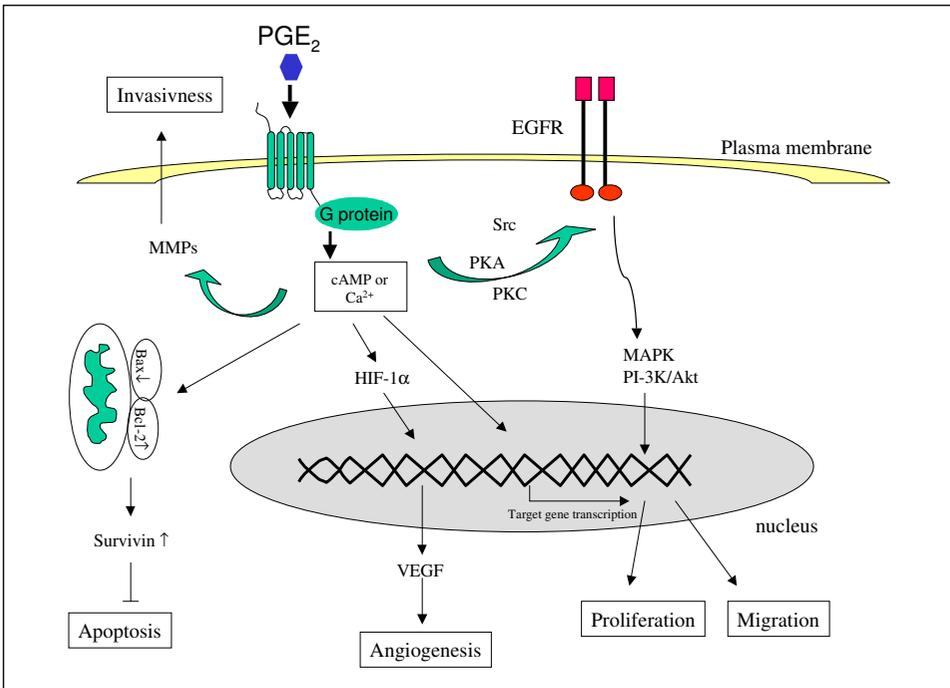


Fig. 2. Different mechanisms by which COX-2 derived prostaglandins are involved in the carcinogenesis.

abnormal survival of cells and the increased risk of mutagenesis and oncogenesis. Apoptotic cell death is characterized by two major pathways, the death receptor pathway and the mitochondrial pathway. The first pathway is triggered by the death receptor superfamily, such as Fas (CD95 receptor and tumour necrosis factor alpha). Binding of Fas-ligand to Fas receptor induces receptor clustering and intracellular recruitment of the adaptor protein, Fas associated protein with death domain (FADD). FADD in turn recruits caspase-8 through the death effector domain, which then activates downstream effector caspases triggering cell death. In the mitochondrial pathway, release of cytochrome c from mitochondria is a key stage, which is controlled by the bcl-2 family of proteins. These can be divided in antiapoptotic members bcl-2, bcl-XL, bcl-w and proapoptotic members bax and bak (13).

COX-2-derived PGs regulate programmed cell death and reduce the apoptotic rate *via* inhibition of the mitochondrial apoptotic pathway characterized by reduced cytochrome c release, attenuated caspase-9 and -3 activation and up-regulation of bcl-2 (14). Additionally, increased prostanoid generation due to COX-2 overexpression specifically inhibits Fas-mediated apoptosis (15). Another evidence supporting the role of PGs in the regulation of apoptotic rate of tumor cells are the studies demonstrating that COX-2 overexpression in these cells

increase their resistance to apoptosis (16). Conversely, COX-inhibitors trigger both the mitochondrial and death receptor-mediated apoptotic pathways with resultant cytochrome c release. In addition, some COX-independent effects on apoptosis have been observed such as inhibition of NF κ B signaling via inhibition of I κ B kinase B activity and by binding to nuclear receptors PPAR (17).

Prostaglandins and increased invasiveness

Tumour cell invasion is an extremely important factor for formation of solid tumours and necessary for their spread to distant organs. COX-2 derived PGs play an important role in the increased invasiveness of cancer cells. This is supported by both *in vitro* and *in vivo* studies. Using COX-2 transfection in human colon cancer cells, Tsuji *et al* were able to show increased invasiveness of COX-2 overexpressing cells and the biochemical and phenotypic changes were accompanied by increased activation of metalloproteinase-2. All these changes were reversed by specific COX-2 inhibitor (18). In another study, COX-2 inhibitor significantly retarded the liver metastasis in a mouse colon cancer model. One of the important mechanisms by which coxibs suppress tumour invasiveness was the inhibition of matrix metalloproteinases (MMP-2 and MM-9) which are known to facilitate cell invasion and migration by degrading extracellular matrix (19).

Cross-talk between epidermal growth factor receptor (EGFR) and COX-2

Recently, an important evidence for cross-talk between COX-2 derived PGs and EGFR have been found. Pai *et al* demonstrated that COX-2 derived PGE₂ can activate EGFR signaling and thereby stimulate cell proliferation. The mechanism by which it occurs appears to be complex. It includes an increased activation of cAMP/protein kinase A pathway leading to increased expression of amphiregulin, a ligand of EGFR which leads to activation of this receptor with resultant increased EGFR signaling and enhanced DNA synthesis. PGE₂ also has been observed to transactivate EGFR *via* intracellular Src-mediated event (20).

Prostaglandins and angiogenesis

Angiogenesis, the formation of new blood vessels from pre-existing vasculature, is an essential process in the carcinogenesis and metastasis. Neovascularization is regulated by the balance between pro-angiogenic and angiogenesis inhibitors in the local tissue environment. Important pro-angiogenic factors include vascular endothelial factor (VEGF), basic fibroblast growth factor (bFGF), interleukin-8, tumor necrosis factor alpha (TNF α), platelet derived growth factor (PDGF) and COX-derived PGs such as PGE₂ and PGI₂. The link between COX-2 derived PGs and angiogenesis is suggested by studies showing a correlation between COX-2 gene expression and angiogenesis in premalignant tissues and cancer. COX-2 expressing cells *in vitro* were shown to secrete proangiogenic factors and these events were inhibited by selective COX-2

inhibitors and non-selective aspirin. PGE₂ stimulate angiogenesis *via* the transcription factor hypoxia inducible factor-1 (HIF-1 alpha) leading to the induction of VEGF. On the other hand, VEGF stimulates COX-2 expression. The ability of COX-2 and VEGF to influence each other suggests a positive feedback amplification mechanism (21, 22).

Gastrointestinal malignancies, including esophageal, gastric and colorectal cancer constitute a significant percentage of all solid tumours worldwide. In recent years, numerous reports have been published that show a possible involvement of COX-2 derived PG_s in the development of these malignancies.

Prostaglandins and immune response

PGs have ability to regulate the immune system. This is of great clinical importance since immunosuppression correlates with the progression of the neoplastic diseases. Previous studies demonstrated that the expression of COX-2 derived PGs may inhibit the antitumour function of dendritic cells playing a key role in the host immune response against tumour antigens (23). In addition, PGE₂ derived from COX-2 suppresses also the activity of natural killer cells and macrophages. Moreover, PGs (especially PGE₂) have profound effects on the production of cytokines by T cells leading to the enhancement of T helper 2 (Th2) type response. By contrast, PGs inhibit the production of Th 1 cytokines (24). Recent studies have shown that the type 1/2 T-helper (Th 1/2) cell balance is shifted toward a Th2-type immune response by malignancy (25). Thus, the overproduction of COX-2 derived PGs could result in the inhibition of cell-mediated antitumour response.

COX-2 and Barrett's esophagus and Barrett's adenocarcinoma

Emerging epidemiological data indicate that PGs may play a pro-oncogenic role in the development of Barrett's adenocarcinoma. Previous studies have demonstrated

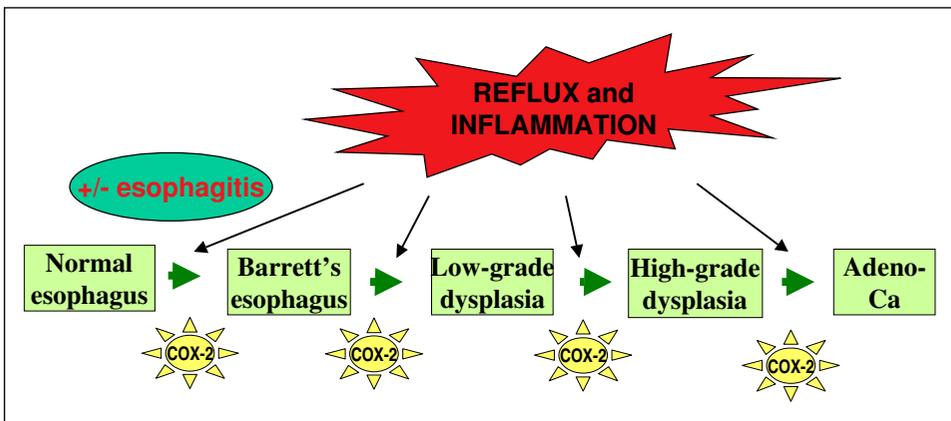


Fig. 3. Schematic presentation of the development of Barrett's carcinoma

that patients taking aspirin have a significantly lower risk for development of both adenocarcinoma and squamous carcinoma of the esophagus (26).

Barrett's esophagus (BE) is defined as the metaplastic conversion of normal esophageal squamous epithelium into columnar intestinalized epithelium with the development of goblet cells. BE is a premalignant condition that may progress to adenocarcinoma called Barrett's carcinoma. Over the past three decades we have observed dramatic increase in the incidence of Barrett's carcinoma while the incidence of esophageal squamous carcinoma remains unchanged. Esophageal adenocarcinoma represents the fastest growing cancer in the western world (27).

Chronic gastroesophageal reflux disease is the most significant risk factor for the development of BE besides white race and male gender (28). Neoplastic progression in BE is a multi-step process in which the metaplastic columnar epithelium sequentially evolves through a metaplasia-dysplasia-carcinoma sequence (*Fig. 3*). This multi-step carcinogenesis in the BE is associated with numerous factors responsible for cancer development such as increased cell proliferation, abnormal expression of growth factors and oncogenes, aberrations in cell cycle control, changes in the expression of adhesion molecules and chromosomal abnormalities (aneuploids) (29). Over the past years, an accumulating evidence has been obtained that increased COX-2 expression could be responsible for chronic inflammation-related cancer promotion (30, 31). Shirvani *et al* demonstrated that the expression of COX-2 increases parallel to the grade of dysplasia observed in BE. Moreover, the same group demonstrated in *ex vivo* model that both gastric acid and bile significantly elevated the expression of COX-2 (32). Further support for the role of COX-2 derived PGs in the carcinogenesis emerged from the animal study by Buttar *et al* (33) in which both non selective COX inhibitor (sulindac) and selective COX-2 blocker (MF tricyclic) significantly attenuated the cancer incidence of Barrett adenocarcinoma. Moreover, Kaur *et al* (34) studied the effect of COX-2 inhibitor (rofecoxib) on the marker of cell proliferation PCNA. In BE the authors found a significantly increased PCNA expression as compared to normal esophageal mucosa. Therapy with rofecoxib caused a significant inhibition of cell proliferation as evidenced by the decreased PCNA expression. Rofecoxib therapy led also to significant downregulation of COX-2 expression in the Barrett's epithelium. Based upon these results, the authors postulated that both biliary and acid reflux cause an increased COX-2 expression with resultant increase of PG synthesis. Increased PG synthesis causes stimulation of cell proliferation and contributes to the development of dysplasia in Barrett's epithelium. Finally, COX-2 expression might be a prognostic marker in patients with Barrett's adenocarcinoma, as expression of COX-2 correlates with patients survival.

COX-2 and gastric cancer

Gastric cancer is one of the most frequent malignancies worldwide (35). PGs play an important role in the gastric carcinogenesis, since the epidemiologic

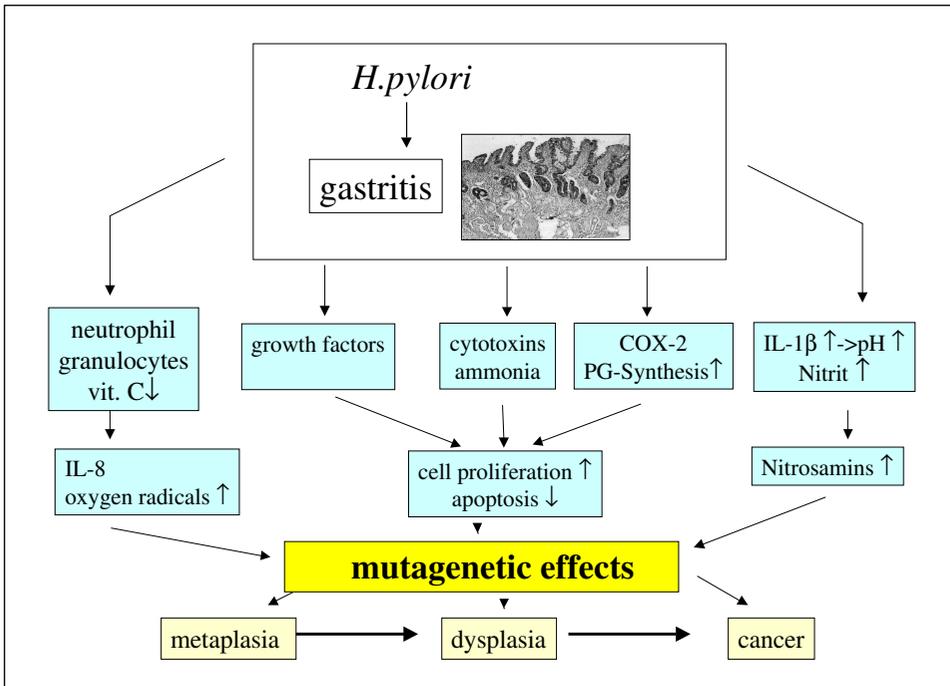


Fig. 4. Role of *H. pylori* in the gastric carcinogenesis.

studies demonstrated that subjects taking regularly aspirin have significantly reduced risk for gastric carcinoma (36). The development of gastric cancer, at least of intestinal type, occurs at the basis of metaplasia-dysplasia-sequence. However, in contrast to esophageal carcinoma, the most important pathophysiological role in the development of gastric cancer plays chronic infection with *H. pylori* (Fig. 4) (37-39). The colonisation of gastric mucosa with this bacterium causes a chronic inflammatory reaction with increased production of proinflammatory cytokines and generation of reactive oxygen species. In addition, *H. pylori*-induced chronic gastritis is associated with an overexpression of COX-2 and increased production of eicosanoids, especially PGE₂ (40). Noteworthy, successful eradication of *H. pylori* infection leads to the significant reduction in COX-2 expression (41).

The importance of COX-2 derived PGs highlight the fact that the expression of this inducible enzyme is highly upregulated in the gastric cancer tissue and this is accompanied by the increased generation of PGs. In addition, COX-2 overexpression enhances the possibility of invasion and metastasis and correlates with a poor prognosis (42,43). Finally, there is also an evidence from animal studies showing a reduced gastric cancer incidence under suppression of COX-2 using specific COX-2 inhibitors (44, 45).

The precise mechanisms leading to the overexpression of COX-2 are still not fully understood. There is evidence that proinflammatory cytokines and

different gastric mucosal growth factors such as transforming growth factor alpha (TGF α) or hepatocyte growth factor (HGF) or finally gastrin could be involved in this process (46).

Especially the interaction between gastrin and COX-2 deserves more attention (Fig. 5). Our previous studies demonstrated an increased gastrin level in the gastric cancer tissue and this was accompanied by the increased release of gastrin in the gastric juice. Moreover, our *in vitro* experiments showed direct and dose-dependent effect of gastrin on the mRNA and protein expression of gastrin in gastric cancer cell lines (46, 47). Gastrin was also a potent stimulator of HGF expression indicating a positive feedback loop between gastrin and gastric mucosal factors. Finally, gastrin possesses also anti-apoptotic capabilities by inducing the anti-apoptotic-proteins Bcl-2 and survivin. The role of gastrin in the gastric carcinogenesis is further supported by the fact that gastric cancer cells are capable of expressing CCK₂ receptor indicating a direct effect of gastrin on these cells (48).

The importance of gastrin and its precursor progastrin in mediating of COX-2 dependent gastric carcinogenesis was demonstrated by our group in humans with gastric cancer treated with COX-2 inhibitor rofecoxib. In this study the median serum progastrin and gastrin levels were found to be significantly higher in gastric cancer patients than in appropriate controls with out gastric cancer (49). Treatment of gastric cancer patients with rofecoxib in a dose 50 mg/day resulted in a significant

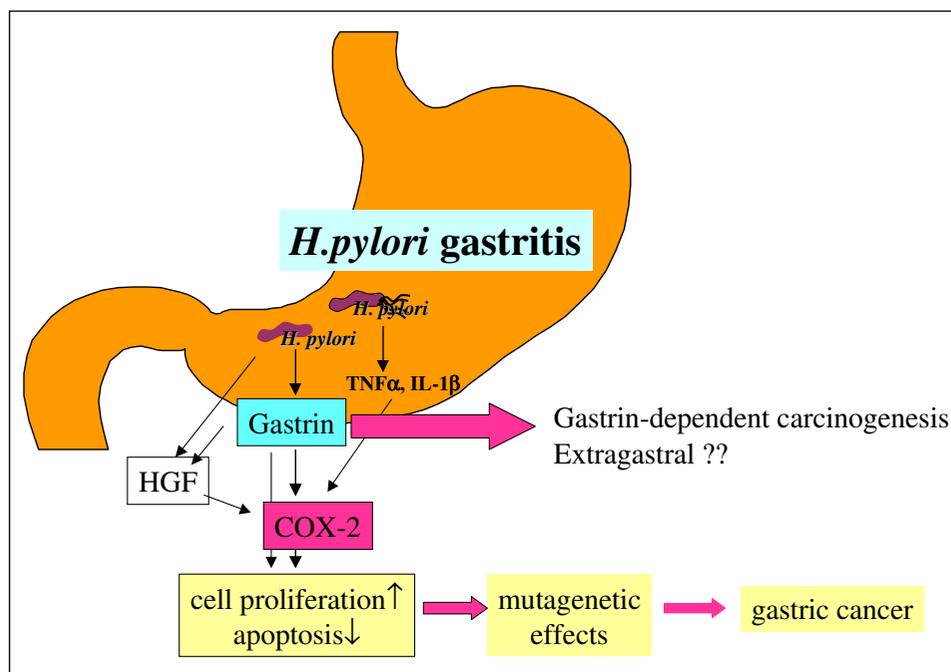


Fig. 5. Gastrin-COX-2 interaction in the pathogenesis of gastric cancer.

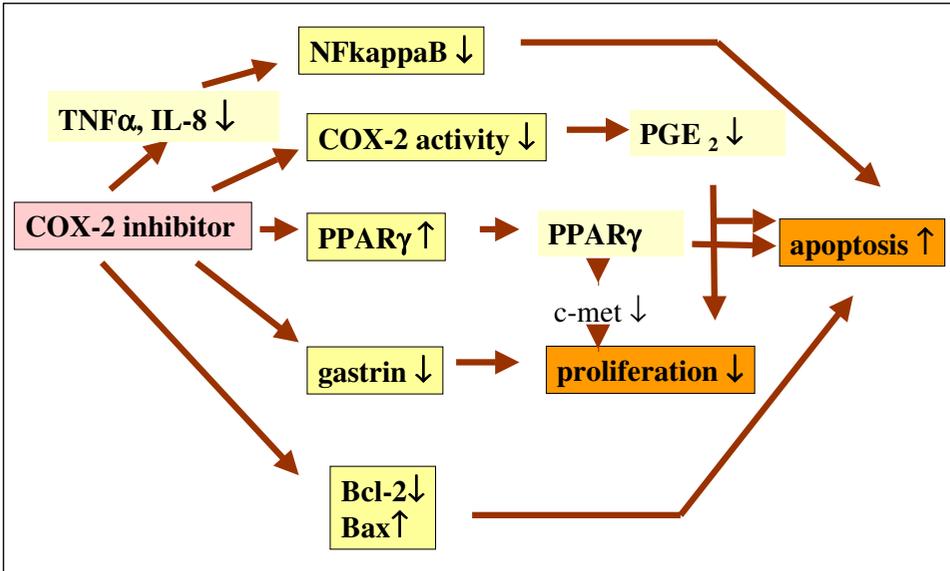


Fig. 6. Different mechanisms of chemoprevention by use of COX-2 inhibitor.

decrease in plasma and tumour contents of both progastrin and gastrin, and this was accompanied by the increased expression of proapoptotic proteins such as Bax and caspase-3 with a concomitant reduction in Bcl-2 and survivin expression. The blockade of COX-2 was also associated with a decrease in the serum level of proinflammatory cytokines IL-8 and TNF α being also involved in the gastric carcinogenesis. Finally, COX-2 inhibition in gastric cancer patients induced in gastric cancer tissue a significant upregulation of peroxisome proliferator activated receptor gamma (PPAR γ), a member of the nuclear receptor superfamily that has been shown to suppress tumorigenesis (Fig. 6). Upon the basis of our results we postulated that the overexpression of PPAR γ might contribute to the limitation of inflammatory process and inhibition of *H. pylori*-induced gastric carcinogenesis (50). The possible mechanisms of chemoprevention induced by COX-2 inhibitor rofecoxib are summarized in the Fig. 7.

COX-2 and colorectal carcinogenesis

Numerous epidemiological studies have demonstrated that the chronic use of aspirin and other NSAIDs is associated with a significantly decreased incidence of colorectal polyps and reduced risk of colon cancer. Although each NSAID has unique physical properties and pharmacokinetics, the mechanism of action common to all NSAIDs is the inhibition of cyclooxygenase activity, especially COX-2 activity (8).

Colorectal cancer (CRC) develops in a stepwise manner from aberrant crypts to adenomas, with increasing grade of dysplasia and finally to cancer. According

to this adenoma-carcinoma sequence model, carcinogenesis proceeds through the accumulation of series of genetic mutations involving several tumour-suppressor genes (APC, p53), oncogenes (k-ras) as well as epigenetic changes (methylation) (51). The previous studies demonstrated that COX-2 is expressed early during this sequence, suggesting an important role in the colorectal carcinogenesis (52). This hypothesis has been strengthened by numerous animal chemoprevention studies. Generally 3 types of models have been used: 1) chemically induced CRC in rats; 2) mice with dysfunctional APC and 3) nude mice subjected to tumour xenografts. In chemically induced CRC in rat, colorectal neoplasms were induced by chemical agents such as 1,2-dimethylhydralazine (DMH) and azoxymethane (AOM). In both models, the inhibition of PGs by non-selective NSAIDs as well as coxibs significantly reduced formation of aberrant crypts and development of adenomas and CRC (53). Moreover, coxibs (rofecoxib) and non selective NSAID (sulindac) reduced significantly the number and size of intestinal polyps in the mice with dysfunctional APC gene (APC Δ^{716} mice) (54). The fact that COX-2 and not COX-1 is overexpressed in the polyps indicate that the activation of COX-2 should be in first line linked to the colorectal carcinogenesis. However, there are some groups postulating that both COX-isoforms are involved in the intestinal tumorigenesis. Chaluda *et al* (55) demonstrated that deficiency of either COX-1 and COX-2 caused similar reduction in intestinal tumorigenesis in Min/+ mice having a mutation in the APC gene and spontaneously developing intestinal

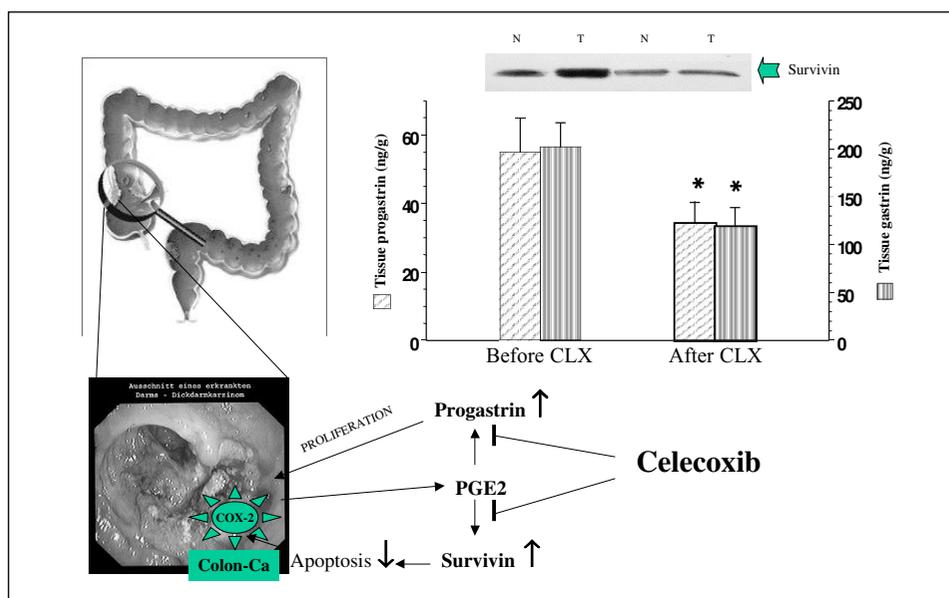


Fig. 6. Effects of celecoxib administration on CRC tissue content of progastrin and gastrin and survivin in CRC patients.

adenomas. Furthermore, both COX-isoforms contributed to PGE₂ production in polyps. Finally, the inhibitory effect of non-selective NSAIDs and coxibs was demonstrated in xenograft mice models in which colorectal cancer cell lines are injected and form tumours with metastasis (56).

Based upon these findings, double-blind randomized placebo-controlled studies were conducted in patients with familial adenomatous polyposis (FAP). FAP is a rare inherited disease caused by a germ-line mutation in the *APC* gene resulting in a dysfunctional APC protein responsible for 1% of CRC detected in the general population. In the study by Steinbach *et al*, six months of twice daily treatment with 400 mg of celecoxib (coxib) led to a significant reduction in the number of colorectal polyps (57). Another placebo-controlled study with another coxib, rofecoxib given daily at a dose 25 mg, demonstrated a significant reduction of rectal polyposis in patients with FAP after 9 months (58). Finally Phillips *et al* showed a significant reduction in duodenal polyps in patients with FAP treated with selective COX-2 inhibitor (59). The data obtained from FAP patients encouraged the conduction of further studies with coxibs in patients with sporadic adenomas and CRC. Barron *et al* investigated the adenoma recurrence in patients with a history of sporadic colorectal adenomas. In this study 1121 patients were randomized to receive placebo (n=372), 81 mg aspirin (n=377) or 325 mg of aspirin (n=375) daily. Follow-up colonoscopy were performed at least 1 year after randomization. Relative risks for advanced lesions were 0.59 (0.38-0.92) in the 81 mg group and 0.83 (0.55-1.23) in 325 mg group as compared to placebo. Surprisingly, the lower aspirin dose had stronger chemopreventive effect than the higher one (60). However, the assessment of possible chemopreventive effect of aspirin on colorectal carcinogenesis was limited by the short follow-up time of the study. The Approve trial (randomized trial of rofecoxib to prevent colorectal adenomas) was a randomized multicenter, placebo controlled, double blind trial to investigate whether the chronic use of the coxib (rofecoxib 25 mg daily) would reduce the adenoma recurrence in patients with a history of colorectal adenomas. 2586 patients were enrolled in this study and randomized to placebo (n=1299) or rofecoxib (n=1287). Therapy with rofecoxib was associated with a significant reduction in adenoma number and size. Unfortunately, an increase in rofecoxib-associated cardiovascular adverse events beginning at 18 months was also noted, which led to early study termination (61).

The role of COX-2 inhibitors in the treatment of advanced human colorectal cancer has not been well defined. Recently, we investigated the effect of celecoxib (CLX) therapy (2x200 mg/day) on the tumour levels of progastrin and gastrin as well as the expression of anti-apoptotic protein Bcl-2 in the tumour tissue and non-tumorous surrounding mucosa in patients with histologically proved CRC. The 14-day therapy with CLX caused a significant decrease in the progastrin and gastrin levels in the CRC tissue as well as significant decrease in the survivin expression (*Fig. 7*). Based upon these results we hypothesize that CLX therapy could contribute to the treatment of CRC *via* suppression of the

anti-apoptotic proteins and reduction in progastrin-promoted tumor growth. The gastrin hypothesis is further supported by the *in vitro* studies performed by Colucci *et al* (62) who demonstrated a direct stimulatory effect of gastrin-17 (G-17) on cell growth and DNA synthesis in colorectal cancer cell line expressing gastrin receptor. In addition, the growth promoting effect of G-17 was accompanied by increased production of PGE₂ and was completely abolished by gastrin receptor antagonists. Moreover, G-17 enhanced the transcriptional activity of COX-2 promoter and stimulated COX-2 expression (63). However, the precise role of gastrin-COX-2 interaction should be investigated in larger human studies.

The combination of chemotherapy with coxibs seems to be an attractive strategy to enhance the antitumour activity since the overexpression of COX-2 in tumor may counteract the efficacy of cytotoxic chemotherapy due to the apoptosis resistance (64-66). The number of clinical studies in which rofecoxib was administered with chemotherapy in patients with CRC is very limited. Beccera *et al* (67) reported a phase II study in which rofecoxib was administered in combination with 5-FU and leucovorin in patients with metastatic CRC. The study was terminated when it was noted an increased toxicity (upper gastrointestinal bleeding, stomatitis, thrombocytopenia, diarrhea) in patients treated with chemotherapy and rofecoxib. The addition of COX-2 inhibitor to the chemotherapy did not increase the efficacy of the antitumour activity of the chemotherapy. Despite these disappointing results, further studies with chemotherapy and COX-2 inhibitors will be needed to determine whether specific COX-2 therapy is able to improve patient outcome with a reasonable safety profile.

We conclude that 1) COX-2 derived PGs play a key role in the tumorigenesis in the gastrointestinal tract; 2) The tumor-promoting effect of PGs is due to their ability to stimulate cell proliferation and migration, inhibit the apoptosis and increase angiogenesis; 3) Increased generation of PGs in the tumor tissue is mainly due to the overexpression of COX-2, but there is also evidence that COX-1 derived PGs could be involved in the carcinogenesis; 4) selective COX-2 inhibitors have been shown to have strong chemopreventive effect on the development of gastrointestinal cancers, however their antitumor efficacy is limited by their cardiovascular adverse effects and 5) selective COX-2 inhibitors due to their anticancer activity represent an interesting alternative treatment for combination therapy with standard chemotherapy in patients with CRC.

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