Aspirin and the Potential Role of Prostaglandins in Colon Cancer

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The arachidonic acid cascade generates a family of bioactive lipids that modulate diverse physiological and pathophysiological responses. Although there has been a great deal of interest in the involvement of arachidonic acid metabolites in cancer, there is uncertainty about which metabolites are the most important and how they contribute to specific steps in cell transformation, tumor growth, and metastasis (1–9). The complexity of arachidonic acid metabolism is daunting to neophytes and the multiplicity of responses exerted by a single metabolite can be confusing to the cognoscenti. This has made it difficult to extrapolate results from animal experiments to human beings.

A recent report from Thun et al. (10) and previous reports from Kune et al. (11) and Rosenberg et al. (12) suggest that aspirin intake may reduce colon cancer incidence (11, 12) and mortality (10) in the general population. Other studies from Paganini-Hill et al. (13, 14) do not support such an association. Aspirin is a relatively specific inhibitor of one branch of the arachidonic acid cascade and even though there is not universal agreement among these epidemiological investigations, their findings refocus attention on the role of arachidonic acid metabolites in cancer. This Perspective was written to highlight some of the existing literature on the aspirin-inhibitable branch of the arachidonic acid cascade and cancer, to apply that literature to colon cancer where possible, and to identify deficiencies in our knowledge that need to be addressed. Although the major focus is the colon, the mechanistic concepts should be applicable to other organ sites.

The Perspective begins with an overview of arachidonic acid metabolism, proceeds to a consideration of the biochemistry and molecular biology of the enzyme inhibited by aspirin, touches on the signal transduction pathways linked to arachidonic acid metabolism, and then considers mechanisms by which specific arachidonic acid metabolites participate in the carcinogenic process. The review is not meant to be comprehensive and apologies are made in advance to colleagues whose work is not highlighted or was inadvertently omitted.

Arachidonic Acid Metabolism

Arachidonic acid is an essential fatty acid consumed in the diet or derived from elongation and desaturation of dietary linoleic acid (15). Most polyunsaturated fatty acids in cells are esterified to phospholipids so release of substrate from these intracellular stores is an important control point in eicosanoid biosynthesis (16, 17). Arachidonic acid mobilization is linked to signal transduction pathways that result in the activation of phospholipase C and phospholipase A2 (18–20). Once arachidonic acid is released from phospholipids it is oxidized by one of three different oxygenases (Fig. 1) (21, 22). Certain cytochromes P-450 insert a single oxygen atom into the double bonds of arachidonic acid to produce epoxy-arachidonic acids (23). Lipoxigenases introduce one molecule of O2 into the carbon framework of arachidonic acid to produce a series of isomeric hydroperoxy acids (24). These hydroperoxy fatty acids are converted to hydroxy fatty acids, to leukotrienes, and to lipoxins (22). Cyclooxygenase introduces two molecules of O2 into arachidonic acid to form the hydroperoxy endoperoxide PGG2, which is reduced by the peroxidase activity to the hydroxy endoperoxide, PGH2 (Fig. 1) (25–27). PGH2 is transformed to PGs, TXA2, PGI2, and MDA (21).3 Oxygenation by cytochromes P-450, lipoxigenases, and cyclooxygenase competes with reesterification to phospholipid; therefore eicosanoid biosynthesis is usually pulsatile (28). Eicosanoids are not stored but are synthesized on demand; therefore inhibitors of the various oxygenases and peroxide-metabolizing enzymes have instantaneous effects on steady-state eicosanoid levels (16, 17).

Each prostaglandin has its own range of biological activities; therefore the physiological response to arachidonic acid oxygenation in a given tissue is largely determined by the levels of PGH2-metabolizing enzymes in the cells making up that tissue (29, 30). For example, platelets divert most of the PGH2 they biosynthesize to TXA2, which is a potent proaggregatory and vasoconstrictive agent (27). In contrast, vascular endothelial cells convert PGH2 mainly to PGI2, which is an inhibitor of platelet aggregation and a vasodilator (31). Potent inhibitors of TXA2 synthase have been developed but selective inhibitors of the other enzymes of PGH2 metabolism are not available (32, 33). Thus, the most common strategy for pharmacological modulation of this branch of the arachidonic acid cascade is inhibition of the cyclooxygenase reaction.

Prostaglandin Endoperoxide Synthase

The protein that oxygenates arachidonic acid to PGG2 also reduces PGG2 to PGH2 (34–36). For this reason, it is called prostaglandin endoperoxide synthase or PGH synthase.4 PGH synthase is a membrane-bound hemeprotein that is widely distributed in mammalian tissue but not present in all cell types (37). The best substrates for its cyclooxygenase activity are 5,8,11,14-eicosatetraenoic acid (arachidonic acid) and 8,11,14-eicosatrienoic acid (17, 38). Other polyunsaturated fatty acids, e.g., 5,8,11,14,17-eicosapentaenoic acid, are not oxygenated as rapidly and under certain conditions act as competitive inhibitors of the oxidation of arachidonic acid (39). The fatty acid composition of cells is sensitive to diet. Arachidonic acid is

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2 The abbreviations used are: PC, prostaglandin; NSAIDs, nonsteroidal antiinflammatory drugs; MDA, malondialdehyde; FANFT, N-[4-(5-nitro-2-furyl)-2-thiazolyl]formamide; PDGF, platelet-derived growth factor; EGF, epidermal growth factor; IGF-1, insulin-like growth factor I; TPA, tetradecanoylphorbol acetate; RPA, retinoylphorbol acetate; ODC, ornithine decarboxylase; LLC, Lewis lung carcinoma; TXA2, thromboxane; PGI2, prostacyclin.

3 For the sake of brevity, PGE2, PGF2α, PGD2, PGI2, and TXA2 will be collectively referred to as prostaglandins even though TXA2 does not contain the prostanoic acid nucleus. The term eicosanoids will be used for all arachidonic acid metabolites including those produced via the action of lipoxigenases.

4 Prostaglandin endoperoxide synthase (EC 1.14.99.1) is also referred to as PGH synthase.

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entaenoic acid can become the most abundant cyclooxygenase acid. HETES, hydroxy fatty compounds produced via the cyclooxygenase pathway.

The sizes of the sheep, human, and mouse PGH synthase genes are approximately 25 kilobases long and are composed of 11 exons (44, 45). The mouse and human PGH synthase genes are approximately 90% similar between mice, and mouse 3T3 cells have been cloned, sequenced, and expressed in heterologous expression systems (41-44). The protein sequences are approximately 90% similar between mice, humans, and sheep. Comparison of sequence homologies and active site labeling experiments have localized the heme binding site and identified potential fifth and sixth ligands to heme iron (44, 45). The mouse and human PGH synthase genes are approximately 25 kilobases long and are composed of 11 exons (Fig. 2) (46, 47). The larger size of the PGH synthase 2 mRNA is due to a much larger 3′-untranslated region (74).

It has been known for some years that prostaglandin synthase in cells is enhanced by a variety of growth factors, tumor promoters, etc. (49, 50). This was thought to be the result of activation of phospholipases and the attendant release of stored substrate. Although phospholipase activation is an important component of the action of certain agonists, many recent studies reveal significant elevation of PGH synthase protein in response to treatment of cells with various agents (51-67). Enhanced expression results from activation at the transcriptional and translational level but the details of the processes are not yet clear (59, 68, 69).

Very recently, three groups reported the detection of a proliferation-associated gene that is homologous to PGH synthase. Transformation of chicken embryo fibroblasts by Rous sarcoma virus causes the stable expression of a protein with approximately 60% similarity to the mouse PGH synthase protein (now termed PGH synthase 1) (58, 70). Likewise, treatment of mouse fibroblasts with TPA, EGF, interleukin 1β, or serum causes transient expression of an immediate early gene that bears approximately 60% similarity to the sheep PGH synthase 1 sequence (71, 72). Both the src- and TPA-stimulated proteins contain many of the key structural elements associated with PGH synthase including the active site residues and the aspirin acetylation site (see below). The complementary DNA for the TPA-stimulated PGH synthase from murine 3T3 cells has been expressed in COS-1 cells and exhibits cyclooxygenase and peroxidase activities (73). This new form of cyclooxygenase from virus-transformed or mitogen-stimulated cells is called PGH synthase 2 and is derived from an 8-kilobase gene composed of 10 exons and 9 introns. The overall exon/intron structure of the 8-kilobase gene is similar to that of PGH synthase 1 (the 25-kilobase gene) although there are significant differences in the regions corresponding to the NH₂ and COOH termini of the proteins (73). In addition, PGH synthase 2 does not contain a sequence corresponding to the exon coding for the hydrophobic leader sequence of PGH synthase 1 (exon B). The mRNA of PGH synthase 2 is approximately 4.1 kilobases long (48).

Inhibition of Prostaglandin Biosynthesis

A trademark of PGH synthase is inhibition of its cyclooxygenase activity by a variety of agents known collectively as NSAIDs (77-80). The best known of these are aspirin, indomethacin, ibuprofen, piroxicam, and sulindac (Fig. 3). Aspirin is a competitive inhibitor and also covalently modifies the PGH synthase protein whereas indomethacin, ibuprofen, piroxicam, and sulindac are competitive inhibitors that do not bind covalently to the protein (44, 81-86). Indomethacin has been demonstrated to form a tight 1:1 complex with PGH synthase that dissociates very slowly (87). Covalent modification of PGH synthase by aspirin is the basis for its irreversible inhibition of cyclooxygenase activity.
Antiinflammatory steroids such as cortisol, dexamethasone, and fluocinolone acetonide also lower prostaglandin production by some cells but they do not inhibit cyclooxygenase activity. Until recently it was believed that steroids inhibited prostaglandin production by inducing the synthesis of a phospholipase inhibitory protein called lipocortin (88). However, this is unlikely to be the major mechanism of inhibition of prostaglandin biosynthesis (89). It has been demonstrated that cortisol and other antiinflammatory steroids are potent inhibitors of interleukin-induced elevation of PGH synthase protein and mechanisms have been proposed at the transcriptional and translational levels to explain this (59, 68, 90, 91). Expression of the PGH synthase 2 gene is particularly sensitive to inhibition by glucocorticoids (48, 92). This may account for the apparent tissue specificity of steroid effects; clinical studies in humans indicate that prostaglandin synthesis in macrophages is particularly sensitive to inhibition by glucocorticoids (93).

**Mechanism of Aspirin Inhibition of Cyclooxygenase Activity**

Aspirin transfers its acetyl moiety to a single serine hydroxyl group of PGH synthase (Ser\(^{530}\)), which blocks the approach of arachidonic acid to the substrate binding site and inhibits cyclooxygenase activity (Equation A) (44, 85)

The acetylated enzyme exhibits normal levels of peroxidase activity (85, 97). Site-directed mutagenesis of the hydroxyl group to a hydrogen atom (Ser\(^{530}\)—>Ala\(^{530}\)) produces an enzyme with undiminished cyclooxygenase activity but no sensitivity to irreversible aspirin inhibition (44). This suggests Ser\(^{530}\) is not essential for cyclooxygenase catalysis and indicates an element of serendipity in the pharmacological activities of aspirin. There are two major determinants of the preferential reactivity of Ser\(^{530}\) to aspirin. The first is the heme prosthetic group (95, 96). Although Ser\(^{530}\) is well removed in linear sequence from the putative heme-binding residues of PGH synthase, acetylation of its hydroxyl group by aspirin is accelerated at least 100-fold by binding heme to the apoprotein. Considering the short half-life of aspirin in humans, this means that it is essentially a titrant for active holoenzyme (95, 97). The other major determinant of acetylation selectivity is the salicylate moiety of aspirin. Reaction of PGH synthase with other acetyl transferring agents does not selectively acetylate Ser\(^{530}\). This suggests that a specific binding site for the salicylate portion of the molecule exists at or near the substrate binding site of the protein. These selectivity determinants account for the fact that PGH synthase is the principal target of aspirin acetylation in intact cells (82).

**Effects of Aspirin on Cardiovascular Disease**

Aspirin acetylation of PGH synthase is irreversible (85). Recovery of cellular cyclooxygenase activity following aspirin treatment requires new protein synthesis. In nucleated cells in culture, this usually occurs in a few hours and is dependent on the presence of growth factors in the culture medium (91, 98). In vivo, aspirin has a half-life of only 5-10 minutes; therefore it does not persist long enough to inhibit newly synthesized protein (97). Cells with low protein synthetic capacity, such as platelets, are unable to recover cyclooxygenase activity. This differential in cyclooxygenase recovery is an important contributing factor to the “selectivity” of aspirin inhibition of platelet cyclooxygenase activity (78). Another contributing factor is pre-systemic acetylation of circulating platelets, which precedes first pass metabolism of aspirin in the liver (99). Thus, TxA\(_2\) synthesis is eliminated by aspirin treatment and does not recover until new platelets are produced (a matter of 3-7 days). In contrast, PGH synthase in vascular endothelial cells is less extensively acetylated than the platelet enzyme and recovers within a few hours after aspirin treatment (100). This results in only a transient reduction in PGI\(_2\) levels. Periodic administration to humans of low doses of aspirin selectively depresses TxA\(_2\) synthesis and reduces platelet responsiveness to agonist stimulation (101-104). A single dose of 40-80 mg aspirin dramatically reduces TxA\(_2\) biosynthesis without significantly affecting PGI\(_2\) biosynthesis (105). This selectivity is lost at doses of 80-325 mg (105). The pharmacological effect of aspirin on the cardiovascular system is an example where the axiom “if a little bit works, a lot works better” does not hold.

Low-dose aspirin was efficacious relative to placebo in preventing myocardial infarction in a selected group of healthy physicians (106). However, the generalizability of this finding to other populations is uncertain. The dose of aspirin used in the Physicians Health Study was equivalent to a single adult aspirin every other day but pharmacokinetic studies suggest that lower doses may be just as effective (107). A timed-release formulation of aspirin appears to maximally depress TxA\(_2\) synthesis without affecting PGI\(_2\) synthesis (108).

**Effects of NSAIDs on Colon Cancer in Animals**

In rat models of colon carcinogenesis, cyclooxygenase inhibitors such as indomethacin, piroxicam, and sulindac exhibit chemopreventive effects as judged by reductions in the number of tumors such as adenomas and adenocarcinomas. These effects are consistent with previously reported studies in colorectal cancer chemoprevention. In this regard, aspirin is unique among nonsteroidal antiinflammatory drugs because it is chemopreventive in both the colon and stomach and has been shown to be efficacious in patients with a history of adenomatous polyps. In contrast, most other NSAIDs are not chemopreventive in the colon and have not been shown to be efficacious in the prevention of adenomatous polyps in humans.

**References**


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![Fig. 3. Structures of some common nonsteroidal antiinflammatory drugs.](image-url)
of tumor-bearing animals and the numbers of tumors per animal (109–117). In the case of piroxicam, a dose-response is observed and there is approximately a 70% reduction in tumor incidence and number at 400 ppm administered in the diet (118). This corresponds to 80% of the maximum tolerated dose of piroxicam. Such a dose of piroxicam might indicate that high doses of cyclooxygenase inhibitors are necessary to inhibit colon carcinogenesis in animals and possibly in humans. However, although piroxicam is a cyclooxygenase inhibitor, it is not an irreversible inhibitor and acts by a different mechanism than aspirin (119). Piroxicam and most of the other cyclooxygenase inhibitors in Fig. 3 may not be as effective as aspirin at selective inhibition of prostaglandin synthesis in individual cell types. Alternatively, they may act by mechanisms independent of cyclooxygenase inhibition (120).

Sulindac has been reported to inhibit dimethylhydrazine-induced colon tumorigenesis in mice when administered in the diet throughout carcinogen administration (117). Both the number of mice with tumors and the number of tumors per animal were reduced. However, if dimethylhydrazine treatment was conducted for 17 weeks before the start of sulindac administration no reduction in tumor growth or development was observed.

**Sulindac and Polyp Regression in Humans**

Waddell and Loughry (121) first reported that sulindac reduces the size and number of rectal polyps in individuals with familial polyposis who have undergone subtotal colectomy and ileoproctostomy. Subsequently, they reported sulindac-induced regression of tumors in affected individuals who had no previous surgical treatment (122). When sulindac administration was discontinued, the tumors recurred and resumption of sulindac treatment caused tumor regression. The reversible nature of sulindac-induced regression was confirmed by Rigau et al. (123).

The results of a randomized, placebo-controlled, double-blind crossover study of sulindac efficacy in a small number of familial polyposis patients were recently reported by Labayle et al. (124). Administration of sulindac at a dose of 300 mg/day (3 × 100 mg) caused complete polyp regression in 6 of the 9 patients and partial regression in the remaining 3 in less than 4 months. Cessation of sulindac treatment resulted in polyp recurrence. The sulindac effect was significant relative to placebo at \( P < 0.01 \). Since the original paper by Waddell and Loughry, 22 cases of polyp regression in familial polyposis patients have been reported. Of these patients, 17 had undergone colectomy with ileorectal anastomosis and 5 had not undergone any surgical treatment. These results imply that sulindac is inhibiting an event in the colon associated with a preneoplastic stage of tumor development. Since the reduced form of sulindac is a cyclooxygenase inhibitor, its ability to induce polyp regression may provide support for the hypothesis that cyclooxygenase inhibition is important to the ability of aspirin to reduce colon cancer mortality (125). In fact, Rigau et al. (123) have demonstrated reduced prostaglandin biosynthetic capacity in colonic mucosal samples taken from patients on long-term sulindac therapy. Another cyclooxygenase inhibitor, indomethacin, did not induce polyp regression in a small number of patients (126). Although this appears at odds with the results obtained with sulindac, it is more likely a reflection of the unusual pharmacokinetics of sulindac. Sulindac itself is not a cyclooxygenase inhibitor but is reduced in vivo to sulindac sulfide which is a powerful cyclooxygenase inhibitor (Equation B) \( d(125, 127, 128) \).

A major site of sulindac reduction is in the colon by bacterial microflora (128, 129). Thus, the produg sulindac is converted to its active metabolite within the colon and achieves relatively high concentrations. Indeed, sulindac and its metabolites are excreted mainly via the intestine (128). In contrast, a small percentage of a dose of indomethacin reaches the colon; most of the indomethacin metabolites are excreted in urine (129, 130). Consistent with the requirement for intracolonic conversion of sulindac to an active metabolite, sulindac does not induce polyp regression in the stomach, small intestine, or ileum (131).

**Aspirin and Human Colon Cancer**

In all four of the epidemiological studies mentioned earlier, aspirin intake was estimated by questionnaire and reported as the frequency of ingestion. In the study by Paganini-Hill et al., daily aspirin intake did not alter the risk of colon cancer whereas in the study by Rosenberg et al., an aspirin intake on 4 or more days per week ("regular use") was associated with a halving of colon cancer incidence (12, 13). In the study by Thun et al. (10), colon cancer mortality decreased with increasing frequency of aspirin use. A frequency of intake of 1–15 times/month was associated with a lower reduction in risk than a frequency of intake of \( \geq 16 \) times/month, which suggests that the effect is dose responsive (10). No frequency of aspirin intake was reported in the study by Kune et al. (11).

In the 3 case-control studies (11–13), aspirin intake was ascertained at the time of colon cancer diagnosis in cases and prior to interview in comparable controls. The larger studies of Kune et al. (\( n = 715 \)) and Rosenberg et al. (\( n = 1326 \)) revealed a halving of colon cancer incidence associated with the use of aspirin (unknown frequency) and regular aspirin or other NSAID use (\( i.e., \) at least 4 times a week for at least 3 months) compared with nonusers of these drugs. In the smaller study of Paganini-Hill (\( n = 181 \) cases) there was actually a 50% increased risk of colon cancer among daily aspirin users that was statistically significant. The cohort study of Thun et al. was the only one that examined mortality from colon cancer (10). Regular aspirin intake at the time of interview was associated with a halving of colon cancer deaths over the next 6 years. None of these studies examined intensity of screening for colon cancer which could be increased among NSAID users. If aspirin use led to bleeding and early detection of polyps or cancers that were subsequently removed, then colon cancer incidence and
mortality could be decreased (132). Thus, although these studies are very provocative, they do not provide sufficient data to accept, at this point, a causal association between aspirin intake and reduced colon cancer incidence and mortality.

Eicosanoids and Signaling

The diverse range of biological activities effected by low concentrations of eicosanoids suggests they are mediators of different signaling pathways. Their rapid metabolic inactivation during pulmonary circulation implies they are not classical circulating hormones but they are likely to have autocrine or paracrine functions (133). As stated earlier, eicosanoids are synthesized on demand in response to cell stimulation by hormones, tumor promoters, membrane perturbation, etc. The rate-limiting step in biosynthesis appears to be release of arachidonic acid from phospholipid stores. This is catalyzed by phospholipase A2 or phospholipase C (20, 134). Many agonists trigger rapid eicosanoid production in cells, which appears to be mediated by phospholipase A2 activation. Arachidonic acid release linked to phospholipase C activation may be a result of hydrolysis of diacylglycerol or the phospholipase C-dependent activation of phospholipase A2 (20). ras transformation of NIH-3T3 cells abolishes prostaglandin synthesis in response to PDGF treatment as a result of reduced phospholipase C activation (135, 136).

Eicosanoid receptors have been reported to be present on many cells including those with eicosanoid biosynthetic capacity (137). Separate receptors have been described for PGE2, PGF2alpha, PGI2, PDG2, and TxA2 (137). It is likely that each eicosanoid has multiple receptor molecules that are coupled to different intracellular signaling pathways (20, 138-150). The TxA2 receptor from platelets has been purified, cloned, and sequenced (151, 152). It appears to be a membrane-spanning, G-protein-linked receptor and there are reports of detection of G-proteins that may be associated with it (142, 148, 153). Likewise, one of the three PGE receptors has recently been cloned and sequenced (154). It also appears to be a G-protein linked receptor.

A good example of eicosanoids acting in an autocrine and paracrine fashion is provided by the regulation of platelet function by TxA2 and PGI2 (155, 156). Stimulation of platelets with low concentrations of thrombin leads to release of arachidonic acid, production of TxA2, and platelet aggregation. TxA2 synthesis and platelet aggregation are inhibited by aspirin. The effects of TxA2 are mediated by its receptor which triggers increases in Ca2+ influx that lead to aggregation. In contrast to the strong platelet-aggregatory effects of TxA2, PGI2 is a potent antiaggregatory agent. PGI2 is a product of arachidonic acid metabolism in vascular endothelial cells and its antiaggregatory effects are mediated by a platelet receptor that is linked to adenylate cyclase activation. Thus, different families of eicosanoid receptors can be present on the same cell and act through different pathways to control cell function. In the case of platelets, one of the eicosanoid mediators is produced in the target cell whereas the other is not. In fact, even though TxA2 is produced in platelets, it is released and stimulates other platelets to aggregate.

The principal role of prostaglandins appears to be as kinetic modulators, either amplifiers or attenuators. Again the platelet provides the best example. TxA2 amplification allows platelets to aggregate at physiological concentrations of thrombin (156). At these low levels of thrombin, aggregation is completely abolished by pretreatment of platelets with aspirin and other cyclooxygenase inhibitors (157). However, cyclooxygenase inhibition does not completely abolish signal transduction pathways leading to aggregation (157). Raising the thrombin concentration overrides the cyclooxygenase block and triggers aggregation by a pathway not sensitive to cyclooxygenase inhibitors. Thus, arachidonic acid metabolism to TxA2 represents only one signal transduction pathway which exists to amplify platelet responsiveness to low thrombin concentrations. PGI2 of vascular origin functions as an attenuator of platelet responsiveness to a wide range of agonists, not just TxA2 (158).

Prostaglandin Synthesis in Human Colon and Colon Cancer

The fact that prostaglandins produced in one cell type can exert an effect on another cell type raises questions about whether the prostaglandins that contribute to tumor development in the colon originate in tumor cells. Numerous studies have demonstrated the ability of human colonic mucosa to synthesize PGE2, PGF2alpha, PDG2, and TxB2 (159-162). In an early study, Bennett et al. (159) reported that the basal level of PGE2 produced by homogenized human colon tumor tissue was higher than that of surrounding normal mucosa. More recently, Narisawa et al. (161) demonstrated that venous blood draining from colon tumors contains large amounts of PGE2 and that the larger the tumor, the higher is the PGE2 output. The levels of PGE2 synthesized by the tumors and present in venous blood correlate with the extent of invasion of surrounding tissue but not with the sites or number of distant metastases. The latter correlate better with the levels of PGE2 in peripheral blood. These observations parallel numerous findings from other laboratories indicating that tumors produce high levels of prostaglandins, most commonly PGE2 (163).

Some cultured tumor cells have been reported to produce high levels of prostaglandins but this observation is by no means universal. Hubbard et al. surveyed 54 different human tumor cell lines for prostaglandin-biosynthetic capacity and found dramatic differences between cell lines from specific anatomical locations (164, 165). The highest levels of prostaglandins were produced by cell lines from non-small cell lung cancers. In nearly all cases, the most abundant products were PGE2 and PGE2alpha (165). Prostaglandins were not synthesized by any of five different small cell lung cancer lines. Of interest for the present discussion, only one of six cell lines derived from colorectal adenocarcinomas and one of three from colorectal carcinomas produced detectable prostaglandins. The levels generated by these two cell lines were among the lowest of all the cell lines surveyed. This sampling suggests that human colon cancer cells are not prodigious producers of prostaglandins especially when compared to cells derived from non-small cell lung cancers.

Maxwell et al. (162) recently confirmed that human colon cancer tissue produces larger amounts of PGE2 than surrounding mucosa. There was not a statistically significant difference in the prostaglandin-biosynthetic capacity of epithelial cells derived from tumor tissue compared to epithelial cells derived from uninvolved tissue. However, a significant difference in biosynthetic capacity was observed between tissue-fixed mononuclear cells derived from tumor compared to uninvolved tissue. Interestingly, no difference in PGE2 synthesis was observed between peripheral mononuclear cells derived from colon cancer patients or normal individuals. The aggregate of these observations suggests that host cells rather than tumor cells may be the major sources of prostaglandins that contribute to colon...
tumorigenesis. This should be borne in mind for subsequent discussions of prostaglandin modulation of cell signaling.

Mechanisms of Malignant Transformation and Growth Linked to Prostaglandin Biosynthesis

Mutagenesis

Malondialdehyde Generation. The development of malignant tumors from human colonic epithelial cells is associated with mutations in oncogenes and tumor suppressor genes (166). The accumulation of genetic alterations appears to be more important than a particular sequence of gene inactivation. This suggests that mutations are an important component of malignant transformation. The cyclooxygenase pathway of arachidonic acid metabolism is responsible for generation of a direct-acting mutagen. MDA is produced by enzymatic and nonenzymatic breakdown of PGH₂ (Fig. 1) (167, 168). MDA is mutagenic in bacterial and mammalian test systems and is carcinogenic in rats (169–172). The enzyme thromboxane synthase is particularly active at MDA production; approximately 50% of the PGH₂ that interacts with TXA₂ synthase is converted to MDA (168). Nearly 100% yields are obtained when endoperoxides derived from other fatty acids (e.g., 8,11,14-eicosatrienoic acid) react with TXA₂ synthase (168).

Another source of MDA is lipid peroxidation (173, 174). MDA production via the cyclooxygenase pathway is inhibited by aspirin whereas production via lipid peroxidation is not. Thus, MDA is an endogenous mutagen and carcinogen produced in eicosanoid-forming tissues, particularly those with high levels of TXA₂ synthase.

MDA induces frame-shift and base-pair substitution mutations in Salmonella typhimurium (169, 175, 176). The frame-shifts induced in the hisD3052 strain of Salmonella have been identified by DNA sequence analysis and are primarily additions (177). The spectrum of mutations induced by reaction of MDA with M13 phage DNA and transformation of Escherichia coli includes frame-shifts and base pair substitutions.⁶ The framemaps are mainly additions and the base pair substitutions are principally G→T transversions and C→T and A→G transitions.⁶ C→T transitions are frequently associated with mutations in the p53 gene in human colon (178). These are presumed to result from deamination of deoxycytidine or 5-methyldeoxycytidine residues but the rate of spontaneous deamination of deoxycytidine is exceptionally slow at physiological pH and temperature (179). The possibility that C→T transitions are induced by reaction of DNA with an endogenous electrophile derived from polyunsaturated fatty acids is an attractive alternative. MDA forms multiple adducts with deoxyguanosine, deoxycytidine, and deoxyadenosine residues (180–184). The extent to which they form in genomic DNA and their role in mutagenesis are uncertain. Likewise, no information is available on the repair of MDA-DNA adducts.

PGH Synthase-dependent Carcinogen Activation. The peroxidase activity of PGH synthase catalyzes the oxidation of a wide range of xenobiotics, including several classes of chemical carcinogens (2, 185–187). Aromatic amines, heterocyclic amines, and dihydrodiol derivatives of polycyclic hydrocarbons are activated to mutagenic derivatives by PGH synthase (188–190). Aspirin inhibits these oxidations by virtue of its ability to prevent cyclooxygenase-catalyzed generation of the hydroperoxide substrate for the peroxidase.

PGH synthase-dependent xenobiotic metabolism has been demonstrated in several intact cellular systems but there is a paucity of data from intact animal experiments. Aspirin administration to rats inhibits bladder tumorigenesis by FANFT and arachidonic acid-dependent oxidation of FANFT by bladder homogenates and cultured urothelial cells (191, 192). These findings indicate a role for PGH synthase in FANFT bladder carcinogenesis. Cyclooxygenase inhibitors have also been reported to inhibit cyclophosphamide-induced pulmonary toxicity and phenytoin-induced teratogenicity (193, 194).

The current consensus is that PGH synthase does not play a major role in systemic xenobiotic oxidation but may be important in toxicity/carcinogenesis in extrahepatic tissues (187, 195). One such tissue is the colon. A survey of 30 different samples of human colon microsomes revealed detectable PGH synthase-dependent oxidation of benzidine in 9 of them (196). In those samples, the extent of benzidine activation by the PGH synthase-dependent pathway was significantly higher than by a mixed-function oxidase-dependent pathway. It was suggested that the interindividual variation in PGH synthase-dependent activation was due to varying degrees of inflammation which remodeled the metabolic capabilities of colonic mucosa by recruitment of cells with high PGH synthase content (196).

Cell Growth

Numerous observations provide a circumstantial link between prostaglandin synthesis and the control of cell growth. As stated earlier, some transformed cell lines synthesize higher amounts of prostaglandins than their nontransformed counterparts and prostaglandin synthesis is stimulated by treatment of these cells with growth factors. Colon tumors cells do not appear to synthesize prostaglandins (164, 165) but prostaglandins derived from other cells in the colonic mucosa or within the tumor may regulate their growth.

PGF₂a stimulates mitogenesis in Swiss 3T3 fibroblasts at concentrations (2 ng/ml) 2–3 orders of magnitude lower than any other prostaglandin (197). PGF₂α-dependent mitogenesis is synergistically stimulated by insulin (198). Basal and insulin-stimulated PGF₂α mitogeneses exhibit parallel sensitivity to alterations in the functional groups in the PGF₂α molecule (199). Of paramount importance are the 9- and 15-hydroxyl groups and the 5,6-double bond (199). These structural determinants mirror those required for binding of agonists to the PGF₂α receptor of ovine and bovine corpora lutea (200, 201).

EGF stimulates synthesis of prostaglandins by BALB/c 3T3 fibroblasts and EGF-dependent proliferation of these cells is inhibited by indomethacin (202). Indomethacin inhibition is overcome by addition of prostaglandins, and PGF₂α is the most potent prostaglandin based on dose response (202). Prostaglandins, including PGF₂α, are not directly mitogenic in these cells but appear to be permissive for EGF mitogenesis. Interestingly, prostaglandins do not appear to be involved in PDGF-dependent mitogenesis of BALB/c 3T3 fibroblasts (203).

PGF₂α also stimulates proliferation of osteoblastic MC3T3-E1 cells (204). The PGF₂α effect is inhibited by antibodies to IGFI-1, which is an autocrine growth factor secreted by the cells. PGF₂α does not enhance secretion of IGF-I but increases the number of high affinity binding sites for IGF-I on the cells (204). Thus, the ability of PGF₂α to stimulate proliferation of MC3T3-E1 osteoblasts appears to be due to its ability to enhance the synthesis of receptors for an autocrine growth factor.

TxA₂ also stimulates cell growth (205, 206). It has been recently demonstrated that a stable TxA₂ analogstimulates proliferation of a rat mammary epithelial cell line and that proliferation is inhibited by a TxA₂ receptor antagonist (207). Administration of the same TxA₂ receptor antagonist to Wistar rats before and after inoculation of tumor cells completely prevents growth of the tumor in vivo. These observations suggest that TxA₂ may be an important stimulus for cell growth and that its effects may be mediated via a receptor. Analysis of the plasma levels of stable metabolites of TxA₂ in breast cancer patients indicates a dramatic decrease following removal of the tumor and an equally dramatic increase coincident with tumor recurrence (208).

A dual role for TxA₂ in regulating human mesangial cell growth in culture has been described (209). Modest stimulation of growth is observed when cells are treated with a stable TxA₂ mimetic agent in the presence of insulin and stimulation is abolished by pretreatment of the cells with a TxA₂ receptor antagonist. However, the same TxA₂ analogue inhibits proliferation of mesangial cells treated with EGF, PDGF, fibroblast growth factor, or fetal bovine serum.

Numerous reports demonstrate that prostaglandins can inhibit proliferation of a wide range of animal and human tumor cells (210–213). Multiple mechanisms of inhibition appear to be involved, one receptor mediated, the other receptor independent (214, 215). The receptor-independent mechanism of inhibition is important for PGE₂ and PGD₂, which can undergo spontaneous or protein-catalyzed dehydration to α,β-unsaturated carbonyl derivatives (Equation C) (216).

\[
\begin{align*}
\text{PGE}_2 & \xrightarrow{\text{H}_2\text{O}} \text{PGA}_2 \\
\end{align*}
\]

The latter derivatives bind to fatty acid-binding protein, accumulate in the nucleus, and induce the synthesis of specific proteins, some of which are associated with the heat shock response (217–223). The antiproliferative effect of these α,β-unsaturated prostaglandins is inhibited by reduced glutathione, which adds to the conjugated double bond (224). The extent to which α,β-unsaturated prostaglandins regulate cell growth in vivo is uncertain. Although their formation requires nonenzymatic dehydration, a dehydrated derivative of PGD₂ has been reported to be present in normal human urine (225).

**Tumor Promotion**

Unraveling the role of prostaglandins in tumor promotion is a perplexing task. The usual problem of multiple levels of eicosanoid involvement is compounded by an incomplete understanding of the molecular events responsible for tumor promotion and differences in compound, tissue, species, and strain sensitivity. Nevertheless, there is strong evidence for prostaglandin involvement in tumor promotion in epidermis of NMRI mice by phorbol esters and this will be considered (226).

A necessary but not sufficient condition for tumor promotion by phorbol esters is hyperplasia, which is preceded by induction of ODC and stimulation of DNA synthesis (227–229). Treatment of CD-1 and NMRI mice with the cyclooxygenase inhibitors indomethacin, aspirin, and flufenamic acid inhibits phorbol ester induction of ODC and mitogenesis (230–232). Inhibition can be overcome by coadministration of PGE₁ or PGE₂ but not PGF₁α or PGF₂α. However, neither PGE₁ nor PGE₂ are mitogens when applied topically to skin (230, 231). Administration of tumor-promoting phorbol esters to mouse skin or cultured murine keratinocytes triggers the release of arachidonic acid and the synthesis of a number of eicosanoids including PGE₂ and PGF₂α (233, 234). A good correlation exists between the inflammatory and tumor-promoting activities of a series of phorbol esters and their ability to stimulate prostaglandin biosynthesis (235). Fractionation of the epidermal cells obtained from SENCAR mice reveals that the highest concentrations of cyclooxygenase and lipoxygenases are present in the differentiated keratinocytes (236). Thus, epidermal cells possess the ability to oxygenate arachidonic acid to prostaglandins. It should be noted that tumor-promoting phorbol esters are highly irritant and trigger the influx of inflammatory cells within 6 h after treatment (237). Inflammatory cells are prodigious generators of eicosanoids; therefore they may contribute to the multiple waves of prostaglandin synthesis that occur in skin following administration of phorbol esters (see below).

Epidermal tumor promotion by phorbol esters has been subdivided into two stages (238, 239). The complete promoter TPA serves as both a stage I and a stage II promoter whereas the incomplete promoter RPA supports only stage II. Administration of one or two doses of TPA followed by multiple doses of RPA reconstitutes a complete promotion response. Topical administration of TPA or RPA triggers two waves of PGE₂ synthesis at 15 and 90 min posttreatment (240). The increase in PGE₂ synthesis at 90 min is accompanied by an increase in PGF₂α synthesis. TPA also induces peaks in PGF₂α synthesis at 3.5–4 and 16 h posttreatment whereas RPA does not. Stimulation of PGF₂α biosynthesis is one of the few biochemical differences between TPA and RPA, which suggests that it may be important for the first stage of promotion. Administration of indomethacin inhibits TPA-dependent stage I promotion but only if it is applied 3 h after TPA, the time at which PGF₂α synthesis is maximal. In fact, coadministration of PGF₂α with indomethacin 3 h after TPA, overcomes the indomethacin-induced block in promotion. The indomethacin block of stage I promotion is not overcome by coadministration of PGE₂. Curiously, promotion by RPA is inhibited by simultaneous administration of indomethacin and this block is also overcome by coadministration of PGF₂α but not PGE₂. Although PGF₂α overcomes the indomethacin block of stage I and stage II promotion, it cannot substitute for TPA or RPA as either a first or second stage promoter (240).

Thus, we are faced with a conundrum. TPA-dependent hyperplasia is essential but not sufficient for tumor promotion and is dependent on the synthesis of PGE₂ in mouse epidermis. However, the biochemical events responsible for stage I and stage II of tumor promotion require the synthesis of PGF₂α. Multiple prostaglandins contribute to promotion; therefore there appears to be a conspiracy of eicosanoids. The highly

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7 TxA₂ has a half-life of 30 s in biological fluids (311). Stable analogues are usually used as surrogates.
probable role of lipoxygenase products in this conspiracy is not considered here but should be noted because lipoxygenase inhibitors abolish phorbol ester tumor promotion in SENCAR and CD-1 mice (241, 242). It is clear that a complex interplay of cell types and mediators contributes to tumor promotion by phorbol esters and non-phorbol ester type compounds. The role of inflammation is not discussed here but should not be ignored because eicosanoids are renowned for their inflammatory, chemotactic, etc., activities which result in the infiltration of a variety of cells that release growth factors, interleukins, etc. Thus, although prostaglandin synthesis is no doubt essential for tumor promotion the precise mechanisms of prostaglandin involvement may be difficult to unravel.

Although most of the studies of prostaglandin participation in tumor promotion have been conducted in mouse skin, some parallel experiments have been conducted in the rat colon using bile acids as promoters. Narisawa et al. (243) have found that indomethacin inhibits the increase in ornithine decarboxylase stimulated by deoxycholic acid by approximately 70%. "Low-to-moderate doses" of PGE2 overcome indomethacin inhibition of ODC induction but do not enhance induction when administered without deoxycholate. Intraepithelial installation of deoxycholate into rats stimulates biosynthesis of PGE2 in the colon and most of the prostaglandin biosynthetic capacity has been localized to the subepithelial layers of the mucosa (244, 245).

Immune Suppression

Another mechanism by which cyclooxygenase inhibitors may reduce tumorigenesis is by modulation of the immune response (246–249). The growth of various tumors is associated with immune suppression in humans and animals (250, 251). One mechanism for immune suppression is dependent on arachidonic acid metabolism. Colony-stimulating factors released by tumor cells activate monocytes and macrophages to synthesize PGE2 which inhibits blastogenesis of T-cells and the cytotoxic activity of natural killer cells (251–254). This appears to be part of a generalized response of the immune system to the presence of foreign antigens (248). Cyclooxygenase inhibitors such as aspirin, indomethacin, piroxicam, and flurbiprofen abolish PGE2 synthesis and attenuate the immune suppression (250, 255–264). This may contribute to their ability to reduce tumor growth in several animal models including the colon.

The effects of cyclooxygenase inhibitors on the growth of the LLC in C57/BL mice have been investigated in some detail. The LLC is a spontaneously arising lung tumor that has been used extensively for screening chemotherapeutic and antimitastatic agents. LLC tumor tissue synthesizes large amounts of eicosanoids and the major product is PGE2 (265). The capacity for PGE2 synthesis is higher in vegetating tissue than necrotic tissue and increases as the tumor grows in vivo (265). The mechanism for increased arachidonic acid oxygenation is unknown but has been suggested to result from increases in PGH synthase activity, induction of phospholipase A2 activity, or infiltration of inflammatory cells (265). The latter possibility seems unlikely because LLC tumors display rather low levels of macrophage contamination and LLC cells release large amounts of PGE2 when purified by centrifugal elutriation or when grown in culture (266, 267).

It is not clear if the high level of cyclooxygenase activity of LLC cells represents an essential feature of malignancy or is a reflection of the pulmonary origin of the LLC (see above). Aspirin and indomethacin significantly inhibit the growth of s.c. LLC tumors but do not inhibit proliferation of LLC cells in culture at concentrations that inhibit PGE2 synthesis (268–271). The ability of indomethacin to inhibit growth of the LLC in vivo correlates to its ability to prevent PGE2-dependent immunosuppression (272–274). Although LLC tumors make considerable amounts of PGE2, the macrophage appears to be the major source of immunomodulatory PGE2 and the principal target for indomethacin (272, 274). Variants of the LLC have been described that exhibit altered morphology when grown in vitro and differing metastatic potential when injected s.c. or i.v. (272). The metastatic variants produce less PGE2 than the nonmetastatic ones but are much more potent at triggering macrophage activation in vivo. This appears to be due to their ability to release colony-stimulating factors (274). Macrophage activation occurs after about 2 weeks of tumor growth and activated macrophages inhibit T-cell proliferation and natural killer cell-mediated cytotoxicity by mixed spleen cell cultures (274). Both effects are abolished by treatment of the macrophages with indomethacin.

Although cyclooxygenase inhibitors reduce the growth of some transplantable tumors, they do not appear to be effective adjuvant chemotherapeutic agents. For example, at longer times after implantation of LLC into C57/BL mice (4 weeks), macrophages lose their immunosuppressive activity and their ability to make PGE2 (275). A second phase of immunosuppression ensues that results from enhanced production of bone marrow-derived suppressors that appear to be monocyte-like (275, 276). This second wave of immunosuppression does not appear to be linked to prostaglandin production because it is not significantly affected by indomethacin treatment of the animals (276). However, its onset is delayed by macrophage PGE2 synthesis which inhibits hematopoiesis (277).

Indomethacin inhibits the growth of s.c. LLC if it is administered several days after the inoculation of tumor cells. However, if further tumor growth occurs before indomethacin administration, it is not an effective inhibitor (269). Thus, cyclooxygenase inhibitors do not appear to be good candidates for treatment of humans with large tumor burdens. Nevertheless, they may slow or prevent the growth of developing tumors.

Considering that a major source of prostaglandins in human colon tumors appears to be tissue macrophages and that macrophage-derived PGE2 appears responsible for immunosuppression, a high research priority is the elucidation of the mechanism of activation of macrophage PGE2 synthesis. Precedents exist for activation and induction of both PGH synthase and phospholipase A2 in macrophages and the recent finding of the PGH synthase 2 gene raises questions about its role in macrophage synthesis of PGH synthase protein.

Finally, it should be noted that although prevention of PGE2-dependent immunosuppression is a plausible mechanism for the antitumor effect of cyclooxygenase inhibitors, indomethacin inhibits the growth of LLC in beige mice and nude mice to the same extent as in normal mice (269). This suggests that T-cells and natural killer cells may not be involved in the antitumor effect of indomethacin against the LLC. Another possibility is that monocytes and macrophages enhance tumor growth by releasing growth factors and that release of these soluble factors is mediated by cyclooxygenase products (269). Alternatively, the indomethacin effect may be independent of cyclooxygenase inhibition (120).
Metastasis

Numerous studies imply a role for eicosanoids in metastasis but there is no consensus on which eicosanoid is responsible or the mechanism(s) by which it acts (278). Indeed, considerable variation is observed in the results of experiments conducted in different model systems. For example, treatment of B16 melanoma cells with indomethacin prior to injection into mice increased the number of metastatic lung colonies in an artificial metastasis model (279, 280). In contrast, treatment of a murine mammary adenocarcinoma with indomethacin prior to injection decreased the number of lung colonies (281). Inhibitory effects of cyclooxygenase inhibitors have also been reported in experiments with several carcinomas and fibrosarcomas (278).

Infusion of pharmacological doses of PGE2, i.v. inhibits metastasis of LLC cells from a s.c. site (spontaneous metastasis) but enhances metastasis of LLC tumor cells tail vein-injected into mice (artificial metastasis) (282, 283). These conflicting results may reflect differences in the events necessary for lung colonization in the two models and their sensitivity to tumor burden. Given the dependence of the outcome on model system, tumor type, cell number, etc., it is difficult to extract mechanistic information that is useful in a general sense.

The importance of prostaglandins, specifically TxA2 and PGI2, in the regulation of platelet function is the basis for a strategy for interrupting the metastatic cascade (284, 285). The formation of tumor cell-platelet aggregates has been proposed to activate tumor cells for attachment to vascular endothelium, assist in the arrest of tumor cells in small capillary beds, or provide an immunological shield for tumor cells (286–288). Furthermore, growth factors released by activated platelets may stimulate growth of the extravasated tumor. On this basis, one might expect antiplatelet agents to inhibit tumor metastasis and a literature documenting this exists (287, 289–293). PGI2, which is a potent inhibitor of platelet aggregation, inhibits metastasis of melanoma, fibrosarcoma, reticulum carcinoma, and carcinomas of lung, breast, colon, pancreas, and prostate (206, 285, 294). Likewise, PGI2 analogues and PGI2 mimetics inhibit metastasis as do agents that inhibit production of TxA2 from PGH2 in platelets (206, 295–297). Although there are some isolated reports indicating a lack of an antimeetastatic effect of PGI2, this may be related to differences in dose, route of administration, preparation of the tumor cells, or metastasis model used (298, 299). In fact, not all tumors stimulate platelet aggregation ex vivo; therefore antiplatelet agents may not be effective antimeetastatic agents in all tumor models (300).

The efficacy of low-dose aspirin as an antiplatelet and anti-thrombotic agent in humans suggests that inhibition of platelet aggregation should be considered as a mechanism for inhibition of colon cancer mortality. Metastatic disease is frequently the cause of death from colon cancer and inhibition of metastasis would certainly have a significant beneficial effect on mortality from colon cancer. Although antiplatelet agents did not enhance survival from an i.m. implanted tumor in an animal model, the relevance of this observation to humans is not certain (297). As discussed earlier, the dose of aspirin estimated from the study of Thun et al. is on the border of low-to-moderate. It is possible that the cell type in which aspirin exerts its preventive effect is highly sensitive to inhibition and slow to recover its cyclooxygenase activity. The cell that best fits this model is the platelet. The mechanistic information that can be inferred from the identification of the cell type inhibited by aspirin reemphasizes the importance of determining the dose response for any protective effects of aspirin on colon cancer mortality.

Conclusion

There is certainly reason to believe that arachidonic acid metabolism contributes to carcinogenesis in humans and that it is possible to modulate its involvement with aspirin or other cyclooxygenase inhibitors. Understanding the mechanisms of action of prostaglandins is complicated by the multistage nature of the carcinogenic process. Considering the available data, it seems to this author that the major contribution of prostaglandins to human colon carcinogenesis is to the growth and possibly spread of the tumor. This opinion is heavily influenced by the reversibility of sulindac inhibition of polyph growth. Of course, for polyphs to develop to cancer additional genetic alterations are required; therefore perhaps mutagenesis by MDA or an electrophile produced by PGH synthase-dependent oxidation plays a role in progression; inhibition of arachidonic acid metabolism could reduce the rate of mutation induction. Since the capacity of arachidonic acid metabolism increases as the tumor grows, so might its susceptibility to mutation.

Assuming for discussion that amplification of cell growth is the key, it is not at all clear which prostaglandins are responsible, where they are made, or how they act. Does the tumor make PGE2α, that up-regulates synthesis of growth factor receptors? Are similar effects exerted by PGE2α, or TxA2 produced by invading macrophages or platelets? Do growing tumors release colony-stimulating factors that activate macrophages to make PGE2 which inhibits proliferation of T-cells and the activity of natural killer cells? Did fewer people die of colon cancer because their aspirin intake rendered their platelets less responsive so that they bled more or had fewer liver metastases? There just isn’t enough information with which to decide.

Many people are already taking an occasional aspirin for prevention of cardiovascular disease and they will probably keep doing so. Should they start taking more? No, experimental studies are needed to confirm the findings of the epidemiological studies, to establish an optimum dose of aspirin, and to determine the benefits and risks of such therapy. The side effects of chronic aspirin intake do not justify ingestion of amounts in excess of that required to elicit the desired pharmacological effect. Other preventive strategies such as reducing fat intake and increasing consumption of fiber, cruciferous vegetables, and fruits may be more worthwhile and are probably safer. In fact, in the study by Kune et al. (11) that first focused attention on aspirin, there was a much more significant protective effect associated with consumption of retinol and vitamin C.

Nevertheless, the possibility that low-dose aspirin might halve colon cancer mortality provides strong impetus for further research in this area. To the extent that the “aspirin effect” reflects cyclooxygenase inhibition, the epidemiological findings reiterate the importance of arachidonic acid metabolism in human cancer and highlight the potential for chemopreventive intervention. Prostaglandins and related compounds are bioactive lipids linked to consumption of dietary fat and they function in intracellular and intercellular signal transduction. Even if they are not obligate intermediates in individual steps in cancer development, they are kinetic modulators of great importance. Actually, the fact that they are not obligate intermediates in signal transduction pathways makes them ideal targets for pharmacological manipulation because inhibition of their
formation should not interrupt basal signal transduction pathways which could prove lethal to normal cells as well as transformed ones. Finally, it is important to reiterate that the pharmaceutical industry has already expended enormous resources on modulating the pathways of arachidonic acid metabolism. Many drugs and drug candidates already exist for which extensive human clinical experience is available. Thus, even if it turns out that aspirin is not the best candidate for an intervention or chemotherapeutic adjuvant trial another cyclooxygenase inhibitor or prostaglandin analogue might be. The opportunity is simply too great to ignore.

This Perspective has concentrated on the involvement in cancer of that branch of the arachidonic acid cascade inhibited by aspirin (the cyclooxygenase branch). It has not considered some of the very exciting recent results implicating the lipooxygenase branch of the cascade in tumor growth and metastasis (301, 302). In fact, there may be a tie between aspirin intake and the lipoxygenase pathway that has not yet been established. The Perspective also did not review studies of dietary fat and colon cancer or animal prevention trials in tissues other than the colon (303–310). All of these topics could and no doubt will be the subject of excellent Perspectives on their own.

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Aspirin and the Potential Role of Prostaglandins in Colon Cancer

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