Am J Clin Oncol (CCT) 26(4 Suppl 2): S48–S57, 2003.

Development of COX Inhibitors in Cancer Prevention and Therapy

Asad Umar, Ph.D., D.V.M., Jaye L. Viner, M.D., M.A., William F. Anderson, M.D., M.P.H., and Ernest T. Hawk, M.D., M.P.H.

On the strength of in vitro, in vivo, observational, and clinical data, nonsteroidal antiinflammatory drugs (NSAIDs)—also referred to as COX inhibitors—have emerged as lead compounds for cancer prevention, and possible adjuncts to cancer therapy. Thus far, the routine use of NSAIDs for these indications is limited, largely owing to toxicity concerns, the paucity of efficacy data for any specific target organ, and uncertainties with regard to the most appropriate regimen (i.e., the best agent, formulation, dose, route of administration, and duration). Strategies to address these concerns primarily aim to improve the therapeutic index (i.e., benefit:risk ratio) of COX inhibitors by 1) minimizing systemic exposures whenever feasible, 2) achieving greater mechanistic specificity, 3) coadministering agents that provide prophylaxis against common toxicities, and 4) coadministering other effective anticancer agents. Clinical trials testing most of these strategies have been completed or are under way. The National Cancer Institute has a substantial research portfolio dedicated to the identification, testing, and development of NSAIDs as preventive and therapeutic anticancer agents. Discovering how to apply NSAIDs in persons with—or at risk for—cancer, although challenging, has the potential for considerable clinical and public health benefits.
Key Words: Cyclooxygenases—COX-1—COX-2—Cancer prevention—Cancer therapy—Molecular target—Clinical trials—Nonsteroidal antiinflammatory drugs (NSAIDs), celecoxib—Sulindac—Aspirin.

Over the last 30 years, we have come to understand that carcinogenesis advances through cumulative structural and functional genomic aberrations that are sequentially expressed within each, and ultimately across all, higher levels of biologic organization (e.g., organelles, cells, tissues, organ).[1] Crucial aberrations may result in a pathologic imbalance between cellular proliferation and apoptosis, thereby fostering a progressively deviant cell population. Fortunately, organisms are endowed with multiple mechanisms—repair, replacement/recruitment, replication, and redundancy—that can preserve structural and functional integrity. Nevertheless, in some instances these mechanisms are overwhelmed and unrepaired damage goes unchecked, potentially leading to the development of cancer.[2]

Understanding the molecular genesis of cancer is integral to advances in cancer prevention and therapy.[3] Technologic innovations in endoscopic and noninvasive imaging, mutation analysis, gene expression, and protein analysis are enhancing our abilities to characterize the cellular and molecular genesis of neoplasia. Data arising from these inquiries are being iteratively transformed into information that can improve cancer risk assessments (i.e., molecular risk profiling), bolster the accuracy and reliability of outcome determinations (i.e., molecular monitoring), and improve anticancer agent identification and development (i.e., molecular targeting).

CYCLOOXYGENASE (COX) AS A MOLECULAR TARGET FOR CANCER PREVENTION AND THERAPY

Ideal molecular targets for anticancer agents are 1) uniquely expressed in neoplasia relative to normal tissues in terms of structure, amount, location, timing, and/or activity; 2) functionally active in initiating or promoting cancer development and progression; 3) accessible to applied interventions; 4) amenable to modulation; and 5) when modulated, result in measurable and reliable clinical benefits (e.g., analgesia; reduced rates of lesion incidence, recurrence, grade; improved quality of life; etc.). Based on these criteria, COX, or perhaps just the COX-2 isoform, is a promising target for both cancer prevention and therapy.

COX activity, which derives from COX-1 and COX-2 isozymes, converts arachidonic acid to PGG_2, and then to PGH_2, which is ultimately metabolized by tissue-specific enzymes in a variety of bioactive prostaglandins (PGs) and thromboxanes. These downstream prostanoids act as auto- or paracrine stimulants, influencing a broad array of physiologic functions. For example, COX-1 produces constitutive prostanoids necessary for normal tissue functions, whereas COX-2 expression is ordinarily low

From the Gastrointestinal & Other Cancers Research Group, National Cancer Institute, Division of Cancer Prevention, Bethesda, Maryland, U.S.A.

Address correspondence and reprint requests to Dr. Ernest T. Hawk, National Cancer Institute, Division of Cancer Prevention, EPN, Suite 2141, 6130 Executive Boulevard, Bethesda, MD 20892-7317, U.S.A.; eh51p@nih.gov

at baseline, but surges in the context of inflammation or neoplasia.[4] Indeed, human neoplasias of most organs—bladder,[5–8] breast,[9,10] uterine cervix,[11,12] central nervous system (CNS),[13–16] colorectum,[10,17–37] esophagus,[38–43] head and neck,[44,45] liver,[46–50] lung,[51–55] pancreas,[56–60] prostate,[61–64] skin,[65–67] and stomach[68–77]—overexpress COX-2 and produce more prostanoids (particularly PGE_2) than healthy tissues from which they are derived. In 1996, Oshima et al.[78] proved the principle that COX-2 was functionally important in carcinogenesis, by demonstrating that knocking out the COX-2 gene in a rodent model of intestinal carcinogenesis resulted in substantial tumor reductions. More recently, investigators have reported the transforming capacity of COX-2 gene transfection in a rodent mammary cancer model.[79] These data confirm the relevance of COX-2 to neoplasia, and the potential importance of COX-2 as a target for cancer prevention and therapy.

NSAIDs as Anticancer Agents

Four complementary lines of evidence establish NSAIDs as important agents for cancer prevention and as possible adjuncts to treatment: 1) COX inhibitors stimulate anticancer effects in in vitro systems; 2) COX inhibitors—and COX-2 gene deletions—inhibit carcinogenesis in carcinogen-induced and genetically driven rodent models (as documented in more than 100 peer-reviewed scientific publications); 3) COX inhibitors reduce the incidence of colorectal precancerous lesions (e.g., adenomas), cancer incidence, and cancer mortality (in more than 25 observational studies); and 4) COX inhibitors regress precancerous lesions (i.e., colorectal aberrant crypt foci and adenomas, and actinic keratoses of the skin) in genetic and sporadic cancer risk cohorts (as reported in more than 17 uncontrolled and controlled clinical trials). The data are most compelling for colorectal neoplasia; however, emerging animal, epidemiologic, or clinical trial data suggest protective and therapeutic effects of NSAIDs in a broad range of extracolonic tissues (e.g., skin, oral mucosa, esophagus, lung, bladder, myeloma, acute myelogenous and chronic lymphocytic leukemias) as well.

In Vitro Data

NSAIDs appear to induce a number of anticancer effects. First, they may serve as competitive inhibitors of COX-related carcinogen activation, thereby rendering threatening exposures less toxic. This mechanism is particularly relevant to the aerodigestive organs (i.e., lung and colon), which are commonly exposed to a broad range of exogenous chemicals (i.e., xenobiotics).[80,81] Cancerous overexpression of COX-2 has also been shown to increase the production of angiogenic growth factors [e.g., vascular endothelial growth factor (VGEF), basic fibroblast growth factor (bFGF), and transforming growth factor-beta (TGF-β)]; and stimulate endothelial cell migration with the formation capillary-like networks.[82] These effects can be blocked by selective COX-2,[82,83] as well as some nonselective COX,[84] inhibitors.

COX-2 overexpression has been clearly associated with suppressed apoptosis. While many molecular pathways regulate apoptosis, BCL-2 family of proteins—and more specifically, the BAX to BCL-2 ratio—is a key determinant of apoptosis. For example, rat intestinal epithelial cells engineered to overexpress COX-2 have decreased amounts of the BAX protein, increased amounts of the BCL-2 protein, and are resistant to apoptosis.[79,85] NSAID inhibition of BCL-2 expression increases the BAX to BCL-2 ratio, thereby restoring apoptosis; human colorectal cancer cells that lack BAX genes have a reduced apoptotic response to NSAIDs.[86] Apoptotic pathways controlled by the Rel/NF-κB family of transcription factors may also be regulated by NSAIDs. Although the exact antiapoptotic mechanisms remain unclear, numerous studies confirm the importance of COX-mediated apoptosis in cancers of the esophagus,[87,88] gallbladder,[89] CNS,[90] head and neck,[91] hematopoietic system,[92] lung,[93] pancreas,[60] and prostate.[94] In total, COX inhibitors appear to reset the balance between cell proliferation and apoptotic cell death, thus prompting tumor stabilization or regression.[95,96]

Chronic inflammation and immune suppression are acknowledged risk factors for epithelial carcinogenesis.[97,98] Inflammation results in the generation of reactive oxygen species and increased prostaglandin E_2 (PGE_2) synthesis, which is in itself immunosuppressive.[99] In contrast, inhibitors of prostaglandin synthesis block immunosuppression and retard tumorigenesis.[100]

Tumor progression is associated with tumor enlargement, lymph node and/or systemic metastases, tumor invasion, and increased cancer-specific mortality. In cancer cell lines, stable overexpression of COX-2 has been associated both with increased production of prostaglandins and increased cellular invasiveness.[101] Consistent with these in vitro findings, selective COX-2 inhibitors have been observed to inhibit metastasis in animals.[102,103] Matrix metalloproteinases (MMPs) and cell adhesion molecules play important roles in tumor invasion, and recent evidence suggests that NSAIDs may modulate both MMPs and cell–cell adhesion.[104]

Animal Data

Twenty-five years of preclinical studies show that inhibition or elimination of COX activity by pharmacologic or genetic manipulations yields fewer animals with tumors, fewer tumors per animal, and smaller tumors.[105] In fact, gene knockout mice carrying a mutant *APC* gene have been used to investigate molecular mechanisms of colorectal polyp formation. When *APC* heterozygotes, the only viable strain of this mutant, were crossed with COX-2 knockout mice, COX-2 +/− mice developed 68% fewer adenomas and COX-2 −/− mice developed 86% fewer adenomas than wild-type COX +/+ mice.[78] Use of pharmacologic COX-2 inhibitors, such as celecoxib or rofecoxib, have also demonstrated profound preventive effects against intestinal carcinogenesis.[106–108] Be-

yond their effects against intestinal cancer, NSAIDs inhibit the development of cancer in a wide variety of organs including the skin,[109] esophagus,[110] lung,[111] breast,[112] prostate,[113] bladder,[114] and hematolymphoid organs.[115,116] NSAIDs also inhibit the growth of a variety of tumor xenografts, suggesting their potential efficacy in therapeutic applications as well.[117]

Human Observational Studies

More than 25 retrospective and prospective epidemiologic studies report that regular aspirin or NSAID use reduces the risk of colorectal adenoma, carcinoma, and/or carcinoma-related mortality by approximately 40% to 50%.[118] These studies show remarkably consistent reductions across the spectrum of colorectal neoplasia regardless of gender, age, tumor location, and familial predisposition. Only two studies have shown no risk reduction for colorectal cancer in persons using aspirin.[119,120]

Although data are less consistent and compelling for extracolonic organs, in aggregate, studies show strong associations between NSAID use and reductions in site-specific cancer risks in the bladder,[121] breast,[122–127] esophagus,[128–131] stomach,[128,132] colorectum (as summarized by Giovannucci),[118] and lung.[126] Indeed, upper GI malignancies in particular seem to be strongly prevented by aspirin and/or NSAID use.[128–133]

Clinical Trials: Nonrandomized and Randomized

In the genetic syndrome of familial adenomatous polyposis (FAP), more than 15 case series or reports describe the chemopreventive effects of sulindac or other NSAIDs against prevalent adenomas.[134] The first report described almost complete regression of colorectal adenomas in four patients given sulindac for 4 to 12 months.[135] Subsequent studies—nonrandomized and randomized, placebo-controlled—have confirmed significant reductions in colorectal adenoma size and number in patients treated with sulindac. Responses to NSAIDs generally occur within a few months, but complete regression is rare and the ramifications of long-term NSAID administration in this cohort have yet to be established.[134] Indeed, four cases of colorectal cancer have been reported in FAP patients while they were taking sulindac, although the implications of these data are difficult to interpret outside the context of larger sulindac-treated cohorts.

With regard to sporadic colorectal neoplasia, the chemopreventive efficacy of NSAIDs has been reported experimentally in three small studies measuring polyp regression, with mixed results.[136–138] The Physicians' Health Study is the only large, placebo-controlled trial of aspirin (325 mg every other day over 5 years) that has been completed to date.[139] At the trial's end, no significant difference was seen in the self-reported frequency of new colorectal malignancies (RR = 1.15; 95% CI = 0.80–1.65).

COX INHIBITORS AND CANCER: THE DHHS RESEARCH PORTFOLIO

The Department of Health and Human Services (DHHS) has a substantial research portfolio dedicated to the identification, testing, and development of NSAIDs and NSAID-derivatives for cancer prevention. The portfolio is detailed in the CRISP (Computer Retrieval of Information on Scientific Projects) Database, a publicly accessible database of biomedical research supported by DHHS, which is updated weekly by the Office of Extramural Research, NIH. A search of this database using keyword combinations of "cyclooxygenase," "nonsteroidal antiinflammatory," "neoplasia," or "cancer" revealed 107 unique projects with 181 associated scientific codes (each project was assigned up to two codes to describe its scientific goals) that were subsequently categorized by primary scientific objective and target organ (depicted in Fig. 1). The portfolio spans a broad range of scientific inquiries, with almost equal representation from the basic and clinical sciences. Included in the portfolio are basic research in key mechanism(s) associated with NSAID efficacy, in vivo NSAID efficacy studies in preinvasive and invasive neoplasia models, and clinical trials testing NSAIDs, NSAID derivatives, selective COX-2 inhibitors, or corticosteroids (which also inhibit COX expression) in different cohorts at risk for cancer of the colorectum, duodenum, esophagus, oral mucosa, lung, skin, or prostate.

IMPROVING THE THERAPEUTIC INDEX OF COX INHIBITORS

Despite persuasive data on the efficacy of NSAIDs against neoplasia/cancer, the development of NSAIDs for preventive and therapeutic anticancer applications has been hampered by concerns relating to potential toxicities, such as gastric ulceration, renal or hepatic dysfunction, and antiplatelet effects. These toxicities are often attributed to COX-1 inhibition, particularly in the context of long-term drug exposure. At least four strategies may improve the therapeutic index of NSAIDs applied with chemopreventive or therapeutic intent.

Reducing Systemic Exposures to NSAIDs

NSAID-induced toxicities are dependent on several factors, including the dose and duration of exposure to the agent, as well as patient age and comorbidities.[140] The therapeutic index of NSAIDs may be improved by topical applications that concentrate NSAID effects on the therapeutic target while minimizing systemic side effects. For example, Wattenberg recently demonstrated the improved efficacy and reduced toxicity of inhalational, as opposed to oral, corticosteroids (e.g., dexamethasone, budesonide) in a carcinogen-induced model of lung neoplasia.[141–143] Based on these findings, topical COX inhibitors are being tested as antineoplastic agents in several NCI-sponsored clinical trials, such as the Phase IIb chemoprevention trial of an aerosolized corti-

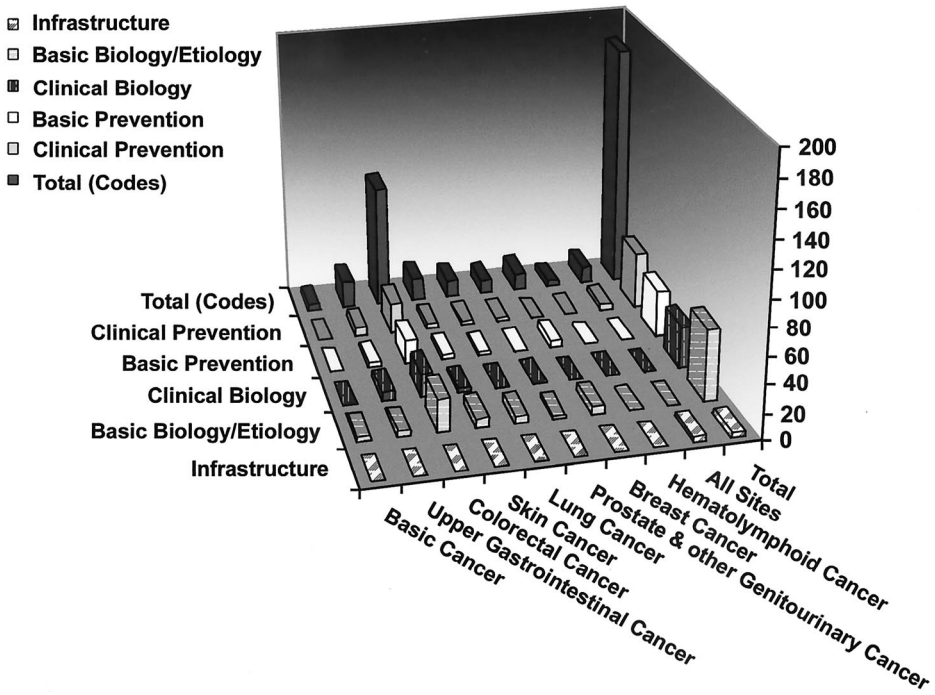

FIG. 1. DHHS Research Portfolio Related to NSAIDs and Cancer of Various Organs, as of 2001. Derived from the CRISP (Computer Retrieval of Information on Scientific Projects), a publicly accessible database of biomedical research supported by the U.S. Department of Health and Human Services. Each contract or grant was assigned up to two codes to reflect the major areas of scientific investigation, as determined by a review of project titles and abstracts.

costeroid in persons with bronchial dysplasia. (S. Lam, personal communication). In addition, the application of topical NSAIDs in cancer prevention was recently given credence by Food and Drug Administration (FDA) approval of topical diclofenac (Solaraze; SkyePharma, Inc.)[144] for the regression of actinic keratoses (http://www.fda.gov/cder/approval/index.htm), which are putative precursors of squamous cell skin cancer.

Dose reduction is another strategy for improving the therapeutic index, and this strategy has been tested in several chemoprevention studies with mixed success. Ruffin et al.[145] conducted a trial of aspirin at doses from 40.5 to 648 mg/d to identify the lowest dose that reliably suppressed PGE_2 concentrations in colorectal mucosa with a minimum of systemic toxicity, and noted that aspirin doses of 81 to 160 mg/d were well tolerated and achieved this goal. These data are tempered by null results from the Physician Health Study, in which aspirin administered at 325 mg every other day over 5 years had no appreciable effect on colorectal neoplasia.[120] Calaluce et al.[146] conducted a Phase IIb trial of piroxicam at 7.5 mg/d over 24 months and noted that although mean PGE_2 concentrations in rectal mucosa were reliably reduced, the incidence of significant GI toxicity was comparable to that seen at higher dose levels; thus the potential for long-term improvement in the therapeutic index with this approach was negligible.

Target Specific Anticancer Activities of NSAIDs

The most striking anticancer properties of NSAIDs (as outlined earlier) include the following: 1) reduction in arachidonic acid products, (2) prevention of free radical–induced genetic damage, 3) interference with the metabolic activation of carcinogens, 4) reduction of proliferation, 5) induction of apoptosis (i.e., restoration of growth regulation in transformed malignant cells), 6) immune stimulation, and 7) antiangiogenic effects.[147] Further inquiry in the specific molecular mechanisms that account for the profound anticancer effects of NSAIDs may facilitate the identification of agents with the most favorable therapeutic indices from among this class of compounds. Although challenging, opportunities to identify new agents with greater mechanistic specificity abound.

NSAID Derivatives

The side effects of traditional NSAIDs, coupled with reported toxicities in approximately 3% of chronic users, motivated the development of derivatives with an improved safety profile that retain the anticancer efficacy of the parent compound. Sulindac sulfone reportedly has negligible COX inhibition and antiinflammatory activity in rodent models, but profoundly reduces tumor incidence, multiplicity, and burden when administered shortly after the induction of carcinogenesis.[148,149] Pub-

TABLE 1. *NCI-sponsored clinical chemoprevention and chemotherapeutic trials with COX inhibitors or NSAID derivatives alone and in combination with other agents**

Intervention	Cohort	Primary Goal	Phase
Prevention trials			
Aspirin	Colorectum—sporadic	Adenoma prevention	III
Celecoxib	Colorectum—FAP	Adenoma suppression	II/III
Celecoxib	Colorectum—HNPCC	Biomarker modulation	II
Celecoxib	Colorectum—sporadic	Adenoma prevention	II
Celecoxib	Colorectum—sporadic	ACF regression	II
Celecoxib	Esophagus—Barrett's	Dysplasia regression/prevention	II
Celecoxib	Prostate	PIN reduction	II
Celecoxib	Bladder	Cancer prevention	II/III
Celecoxib	Skin—sporadic	AK regression/prevention	II/III
Celecoxib	Skin—genetic	Cancer suppression/prevention	II
Sulindac sulfone	Duodenum—FAP	Adenoma regression	II
Aspirin + folate	Colorectum—sporadic	Adenoma prevention	III
Celecoxib + DFMO	Colorectum—FAP	Adenoma regression	III
Celecoxib + selenium	Colorectum—sporadic	Adenoma prevention	III
Celecoxib + selenium	Esophagus—squamous	Dysplasia regression/prevention	II
Piroxicam + calcium	Colorectum—sporadic	Adenoma prevention	III
Sulindac + DFMO	Colorectum—sporadic	Adenoma prevention	II/III
Therapy Trials			
Celecoxib	Prostate	Efficacy study	I
Celecoxib/5-FU/cisplatin/XRT/brachytherapy	Cervical	Efficacy study	I/II
Celecoxib/trastuzumab	Breast—metastatic	Efficacy study in HER2/neu breast cancer	II
Indomethacin/5-FU/phenylbutyrate	CRC—metastatic	Dose escalation study of 5-FU	I/II

Source, NCI's cancer database PDQ (physician's data query).
* As of 2001.
DFMO, α-difluoromethylornithine; FAP, familial adenomatous polyposis; HNPCC, hereditary nonpolyposis colorectal carcinoma; ACF, aberrant crypt foci; PIN, prostatic intraepithelial neoplasia; AK, actinic keratosis; 5-FU, 5-fluorouracil; XRT, external beam radiotherapy.

lished efficacy data from chemoprevention studies of Min mice are less impressive.[150,151] Van Stolk recently conducted a phase I/II trial of sulindac sulfone given over 6 months in patients with FAP in which adenoma number and size were stabilized.[152] Data from other clinical trials in cancer prevention and therapy have yet to be published. *R*-flurbiprofen, a noncyclooxygenase-inhibiting enantiomer of flurbiprofen, has little of the ulcerogenic tendency of the parent compound, flurbiprofen.[153] Nevertheless, preclinical studies of *R*-flurbiprofen have shown substantial reductions in tumor multiplicity and prolonged survival,[113,151,154] although these results have yet to be extended to human chemoprevention trials.

Selective COX-2 Inhibitors

The therapeutic index of NSAIDs improved with the discovery of COX-2 selective inhibitors [e.g., celecoxib (Celebrex, Pharmacia, Inc.) and rofecoxib (Vioxx, Merck and Co.], which recently received FDA approval as antiinflammatory agents, and have subsequently become lead compounds for cancer prevention and therapy. In a recent trial involving more than 8,000 patients with arthritis, celecoxib was associated with 40% to 50% fewer symptomatic ulcers compared to other nonselective COX inhibitors.[155] Thus, COX-2 selective inhibitors have partially fulfilled the search for a "safer" aspirin.[156]

Nine in vivo studies—two involving NS-398, one with MF tricyclic, two with nimesulide, one with rofecoxib, and three with celecoxib—have proven the efficacy of COX-2 selective inhibitors in reducing aberrant crypt foci and colorectal tumors in carcinogen-induced and genetically-induced rodent models.[78,107,108,157–162] In addition, profound preventive effects have been reported with COX-2 selective inhibitors in animal models of breast,[163–165] skin,[166,167] bladder,[168,169] lung,[170] and prostate[94] cancer. Based on these impressive preclinical efficacy data, COX-2 selective inhibitors advanced in clinical cancer prevention and therapy trials. Although one small case series of nimesulide administered over 10 weeks showed no reduction in rectal adenoma burden in seven persons with FAP,[171] a more definitive randomized, placebo-controlled trial of celecoxib administered over 6 months to 83 FAP subjects showed significant regression and reductions in colorectal adenoma number and size, with a frequency and side effect profile comparable to placebo.[172] In the latter study, celecoxib improved the endoscopic appearance of both the colorectum and the duodenum, suggesting that it may reduce FAP-related neoplastic risk in both organs. On the strength of these data, the FDA approved celecoxib as an adjunct to standard surveillance and prophylactic surgery for patients with FAP (http://www.fda.gov/cder/approval/index.htm).

On the strength of preclinical and clinical efficacy data with celecoxib and rofecoxib, and the ubiquitous overexpression of COX-2 in human neoplasia, the NCI is

collaborating with academic and pharmaceutical collaborators on a broad range of Phase II/III cancer prevention and treatment trials with COX inhibitors (Table 1).

Provide Prophylaxis Against Gastrointestinal Toxicities of NSAIDs

A recent Cochrane Review of randomized, controlled clinical trials of agents that provide prophylaxis against NSAID-induced gastropathy suggests that misoprostol, proton pump inhibitors, and double-dose H2-receptor antagonists protect against chronic NSAID-related upper GI endoscopic damage.[173] Coadministration of any of these agents with NSAIDs for cancer prevention has yet to be fully explored, both in animals and humans, to determine whether prophylaxes such as these impair the anticancer efficacy, or the incidence of NSAID-related side effects in persons with, or at risk for, cancer.

Coadminister With Other Anticancer Interventions

Finally, the therapeutic index of NSAIDs may be enhanced through combinations with other effective dietary manipulations, chemopreventives, or chemotherapeutics.[174,175] In carcinogen-induced and genetically driven rodent models of colorectal cancer, for example, the synergistic efficacy of NSAIDs in combination with difluoromethylornithine (DFMO), an inhibitor of ornithine decarboxylase, has been reliably and reproducibly demonstrated.[176–180] In addition, several studies show that the same degree of inhibition is sustained even with 30% to 50% reductions of the NSAID, DFMO, or both. NSAIDs have been shown to boost the preclinical efficacy of other classes of agents as well, including statins and EGFR inhibitors such as EKB-569 (Wyeth-Ayerst).[181,182] This strategy holds tremendous promise for cancer prevention and treatment,[183] and is being actively investigated in NCI-sponsored clinical trials (Table 1).

COX INHIBITORS IN CANCER THERAPY

COX inhibitors are showing promise as adjuncts to cancer treatment. Preclinical studies dating back 15 years demonstrate the efficacy of NSAIDs, and more recently NSAID derivatives and COX-2 inhibitors, in reducing the growth of tumor xenografts including melanoma[184] and cancers of the stomach,[185] colorectum,[186–188] and prostate.[189] Despite these promising findings, the only published clinical efficacy data come from one randomized, placebo-controlled trial of indomethacin or prednisolone in 135 patients with malnutrition and various advanced malignancies.[190] At the conclusion of the trial, Lundholm and colleagues reported that compared to the placebo group, patients taking indomethacin had significantly less pain and somewhat surprisingly, an improved mean survival (510 vs. 250 days) as well. This finding needs independent corroboration before it can be considered definitive, but it provides a compelling impetus for additional work to fully explore the potential of NSAIDs in cancer therapy.

LOOKING TO THE FUTURE: NSAIDs IN CANCER PREVENTION AND THERAPY

The long-term risks and benefits of a preventive or therapeutic agent depend on several factors, including the agent's mechanism(s) of action; pharmacokinetic properties (absorption, distribution, metabolism, and excretion); intended route, frequency, and duration of use; and the clinical risks and options of the intended cohort(s). In aggregate, these complex factors drive the development of promising agents, the utility of which can only be established through rigorous clinical trials that carefully pair agents with appropriate cohorts based on known mechanisms of risk and activity.

NSAIDs and COX-2 selective inhibitors—particularly when combined with other anticancer agents—have enormous appeal because they are relatively safe and could offer efficacy against a broad range of common chronic diseases such as cancer, arthritis, cognitive degeneration, and cataracts.[191–193] As a result, the federal government, industry, and academia are devoting substantial resources to test antiinflammatory drugs for the management of these conditions. If NSAIDs are shown to reduce the incidence of cancer at one or multiple sites, their potential utility is staggering. Indeed, discovering how to apply NSAIDs, COX-2 selective inhibitors, or both in persons at risk for these common diseases of aging has the potential for significant clinical and public health rewards. ℭ

REFERENCES

1. Hawk E, Viner JL, Lawrence JA. Biomarkers as surrogates for cancer development. *Curr Oncol Reports* 2000;2:242–50.
2. Umar A, Kunkel TA. DNA-replication fidelity, mismatch repair and genome instability in cancer cells. *Eur J Biochem* 1996;238: 297–307.
3. Kelloff GJ, Crowell JA, Steele VE, et al. Progress in cancer chemoprevention. *Ann NY Acad Sci* 1999;889:1–13.
4. Vane JR, Bakhle YS, Botting RM. Cyclooxygenases 1 and 2. *Annu Rev Pharmacol Toxicol* 1998;38:97–120.
5. Bostrom PJ, Uotila P, Rajala P, et al. Interferon-alpha inhibits cyclooxygenase-1 and stimulates cyclooxygenase-2 expression in bladder cancer cells in vitro. *Urol Res* 2001;29:20–4.
6. Komhoff M, Guan Y, Shappell HW, et al. Enhanced expression of cyclooxygenase-2 in high grade human transitional cell bladder carcinomas. *Am J Pathol* 2000;157:29–35.
7. Mohammed SI, Knapp DW, Bostwick DG, et al. Expression of cyclooxygenase-2 (COX-2) in human invasive transitional cell carcinoma (TCC) of the urinary bladder. *Cancer Res* 1999;59: 5647–50.
8. Shirahama T, Arima J, Akiba S, et al. Relation between cyclooxygenase-2 expression and tumor invasiveness and patient survival in transitional cell carcinoma of the urinary bladder. *Cancer* 2001;92:188–93.
9. Hwang D, Scollard D, Byrne J, et al. Expression of cyclooxygenase-1 and cyclooxygenase-2 in human breast cancer. *J Natl Cancer Inst* 1998;90:455–60.
10. Soslow RA, Dannenberg AJ, Rush D, et al. COX-2 is expressed in human pulmonary, colonic, and mammary tumors. *Cancer* 2000;89:2637–45.
11. Gaffney DK, Holden J, Davis M, et al. Elevated cyclooxygenase-2 expression correlates with diminished survival in carcinoma of the cervix treated with radiotherapy. *Int J Radiat Oncol Biol Phys* 2001;49:1213–7.

12. Kulkarni S, Rader JS, Zhang F, et al. Cyclooxygenase-2 is overexpressed in human cervical cancer. *Clin Cancer Res* 2001; 7:429–34.
13. Deininger MH, Weller M, Streffer J, et al. Patterns of cyclooxygenase-1 and -2 expression in human gliomas in vivo. *Acta Neuropathol (Berl)* 1999;98:240–4.
14. Joki T, Heese O, Nikas DC, et al. Expression of cyclooxygenase 2 (COX-2) in human glioma and in vitro inhibition by a specific COX-2 inhibitor, NS-398. *Cancer Res* 2000;60:4926–31.
15. Karim MM, Hayashi Y, Inoue M, et al. Cox-2 expression in retinoblastoma. *Am J Ophthalmol* 2000;129:398–401.
16. Shono T, Tofilon PJ, Bruner JM, et al. Cyclooxygenase-2 expression in human gliomas: prognostic significance and molecular correlations. *Cancer Res* 2001;61:4375–81.
17. Chapple KS, Cartwright EJ, Hawcroft G, et al. Localization of cyclooxygenase-2 in human sporadic colorectal adenomas. *Am J Pathol* 2000;156:545–53.
18. Fujita T, Matsui M, Takaku K, et al. Size- and invasion-dependent increase in cyclooxygenase 2 levels in human colorectal carcinomas. *Cancer Res* 1998;58:4823–6.
19. Fujita M, Fukui H, Kusaka T, et al. Relationship between cyclooxygenase-2 expression and K-ras gene mutation in colorectal adenomas. *J Gastroenterol Hepatol* 2000;15:1277–81.
20. Bamba H, Ota S, Kato A, et al. High expression of cyclooxygenase-2 in macrophages of human colonic adenoma. *Int J Cancer* 1999;83:470–5.
21. Eberhart CE, Coffey RJ, Radhika A, et al. Up-regulation of cyclooxygenase 2 gene expression in human colorectal adenomas and adenocarcinomas. *Gastroenterology* 1994;107:1183–8.
22. Hao X, Bishop AE, Wallace M, et al. Early expression of cyclo-oxygenase-2 during sporadic colorectal carcinogenesis. *J Pathol* 1999;187:295–301.
23. Arbabi S, Rosengart MR, Garcia I, et al. Epithelial cyclooxygenase-2 expression: a model for pathogenesis of colon cancer. *J Surg Res* 2001;97:60–4.
24. Chen WS, Wei SJ, Liu JM, et al. Tumor invasiveness and liver metastasis of colon cancer cells correlated with cyclooxygenase-2 (COX-2) expression and inhibited by a COX-2-selective inhibitor, etodolac. *Int J Cancer* 2001;91:894–9.
25. Dimberg J, Samuelsson A, Hugander A, et al. Differential expression of cyclooxygenase 2 in human colorectal cancer. *Gut* 1999;45:730–2.
26. Gustafson-Svard C, Lilja I, Hallbook O, et al. Cyclooxygenase-1 and cyclooxygenase-2 gene expression in human colorectal adenocarcinomas and in azoxymethane induced colonic tumours in rats. *Gut* 1996;38:79–84.
27. Hull MA, Fenwick SW, Chapple KS, et al. Cyclooxygenase-2 expression in colorectal cancer liver metastases. *Clin Exp Metastasis* 2000;18:21–7.
28. Kargman SL, O'Neill GP, Vickers PJ, et al. Expression of prostaglandin G/H synthase-1 and -2 protein in human colon cancer. *Cancer Res* 1995;55:2556–9.
29. Kutchera W, Jones DA, Matsunami N, et al. Prostaglandin H synthase 2 is expressed abnormally in human colon cancer: evidence for a transcriptional effect. *Proc Natl Acad Sci U S A* 1996;93:4816–20.
30. Maekawa M, Sugano K, Sano H, et al. Increased expression of cyclooxygenase-2 to -1 in human colorectal cancers and adenomas, but not in hyperplastic polyps. *Jpn J Clin Oncol* 1998;28:421–6.
31. Masunaga R, Kohno H, Dhar DK, et al. Cyclooxygenase-2 expression correlates with tumor neovascularization and prognosis in human colorectal carcinoma patients. *Clin Cancer Res* 2000;6:4064–8.
32. Sakuma K, Fujimori T, Hirabayashi K, et al. Cyclooxygenase (COX)-2 immunoreactivity and relationship to p53 and Ki-67 expression in colorectal cancer. *J Gastroenterol* 1999;34:189–94.
33. Sano H, Kawahito Y, Wilder RL, et al. Expression of cyclooxygenase-1 and -2 in human colorectal cancer. *Cancer Res* 1995; 55:3785–9.
34. Shattuck-Brandt RL, Lamps LW, Heppner Goss KJ, et al. Differential expression of matrilysin and cyclooxygenase-2 in intestinal and colorectal neoplasms. *Mol Carcinog* 1999;24:177–87.
35. Sheehan KM, Sheahan K, O'Donoghue DP, et al. The relationship between cyclooxygenase-2 expression and colorectal cancer. *JAMA* 1999;282:1254–7.
36. Sinicrope FA, Lemoine M, Xi L, et al. Reduced expression of cyclooxygenase 2 proteins in hereditary nonpolyposis colorectal cancers relative to sporadic cancers. *Gastroenterology* 1999;117:350–8.
37. Tomozawa S, Tsuno NH, Sunami E, et al. Cyclooxygenase-2 overexpression correlates with tumour recurrence, especially haematogenous metastasis, of colorectal cancer. *Br J Cancer* 2000;83:324–8.
38. Kandil HM, Tanner G, Smalley W, et al. Cyclooxygenase-2 expression in Barrett's esophagus. *Dig Dis Sci* 2001;46:785–9.
39. Morris CD, Armstrong GR, Bigley G, et al. Cyclooxygenase-2 expression in the Barrett's metaplasia-dysplasia-adenocarcinoma sequence. *Am J Gastroenterol* 2001;96:990–6.
40. Ratnasinghe D, Tangrea J, Roth MJ, et al. Expression of cyclooxygenase-2 in human squamous cell carcinoma of the esophagus; an immunohistochemical survey. *Anticancer Res* 1999;19: 171–4.
41. Shirvani VN, Ouatu-Lascar R, Kaur BS, et al. Cyclooxygenase 2 expression in Barrett's esophagus and adenocarcinoma: ex vivo induction by bile salts and acid exposure. *Gastroenterology* 2000;118:487–96.
42. Wilson KT, Fu S, Ramanujam KS, et al. Increased expression of inducible nitric oxide synthase and cyclooxygenase-2 in Barrett's esophagus and associated adenocarcinomas. *Cancer Res* 1998;58:2929–34.
43. Zimmermann KC, Sarbia M, Weber AA, et al. Cyclooxygenase-2 expression in human esophageal carcinoma. *Cancer Res* 1999; 59:198–204.
44. Chan G, Boyle JO, Yang EK, et al. Cyclooxygenase-2 expression is up-regulated in squamous cell carcinoma of the head and neck. *Cancer Res* 1999;59:991–4.
45. Gallo O, Franchi A, Magnelli L, et al. Cyclooxygenase-2 pathway correlates with vegf expression in head and neck cancer. Implications for tumor angiogenesis and metastasis. *Neoplasia* 2001; 3:53–61.
46. Bae SH, Jung ES, Park YM, et al. Expression of cyclooxygenase-2 (COX-2) in hepatocellular carcinoma and growth inhibition of hepatoma cell lines by a COX-2 inhibitor, NS-398. *Clin Cancer Res* 2001;7:1410–8.
47. Koga H, Sakisaka S, Ohishi M, et al. Expression of cyclooxygenase-2 in human hepatocellular carcinoma: relevance to tumor dedifferentiation. *Hepatology* 1999;29:688–96.
48. Kondo M, Yamamoto H, Nagano H, et al. Increased expression of COX-2 in nontumor liver tissue is associated with shorter disease-free survival in patients with hepatocellular carcinoma. *Clin Cancer Res* 1999;5:4005–12.
49. Rahman MA, Dhar DK, Yamaguchi E, et al. Coexpression of inducible nitric oxide synthase and cox-2 in hepatocellular carcinoma and surrounding liver: possible involvement of cox-2 in the angiogenesis of hepatitis c virus-positive cases. *Clin Cancer Res* 2001;7:1325–32.
50. Shiota G, Okubo M, Noumi T, et al. Cyclooxygenase-2 expression in hepatocellular carcinoma. *Hepatogastroenterology* 1999; 46:407–12.
51. Achiwa H, Yatabe Y, Hida T, et al. Prognostic significance of elevated cyclooxygenase 2 expression in primary, resected lung adenocarcinomas. *Clin Cancer Res* 1999;5:1001–5.
52. Dohadwala M, Luo J, Zhu L, et al. Non-small cell lung cancer cyclooxygenase-2-dependent invasion is mediated by CD44. *J Biol Chem* 2001;24:24.
53. Hida T, Yatabe Y, Achiwa H, et al. Increased expression of cyclooxygenase 2 occurs frequently in human lung cancers, specifically in adenocarcinomas. *Cancer Res* 1998;58:3761–4.
54. Hosomi Y, Yokose T, Hirose Y, et al. Increased cyclooxygenase 2 (COX-2) expression occurs frequently in precursor lesions of human adenocarcinoma of the lung. *Lung Cancer* 2000;30:73–81.
55. Huang M, Stolina M, Sharma S, et al. Non-small cell lung cancer cyclooxygenase-2-dependent regulation of cytokine balance in lymphocytes and macrophages: up-regulation of interleukin 10 and down-regulation of interleukin 12 production. *Cancer Res* 1998;58:1208–16.

56. Kokawa A, Kondo H, Gotoda T, et al. Increased expression of cyclooxygenase-2 in human pancreatic neoplasms and potential for chemoprevention by cyclooxygenase inhibitors. *Cancer* 2001; 91:333–8.
57. Molina MA, Sitja-Arnau M, Lemoine MG, et al. Increased cyclooxygenase-2 expression in human pancreatic carcinomas and cell lines: growth inhibition by nonsteroidal anti-inflammatory drugs. *Cancer Res* 1999;59:4356–62.
58. Okami J, Yamamoto H, Fujiwara Y, et al. Overexpression of cyclooxygenase-2 in carcinoma of the pancreas. *Clin Cancer Res* 1999;5:2018–24.
59. Tucker ON, Dannenberg AJ, Yang EK, et al. Cyclooxygenase-2 expression is up-regulated in human pancreatic cancer. *Cancer Res* 1999;59:987–90.
60. Yip-Schneider MT, Barnard DS, Billings SD, et al. Cyclooxygenase-2 expression in human pancreatic adenocarcinomas. *Carcinogenesis* 2000;21:139–46.
61. Tanji N, Kikugawa T, Yokoyama M. Immunohistochemical study of cyclooxygenases in prostatic adenocarcinoma; relationship to apoptosis and Bcl-2 protein expression. *Anticancer Res* 2000;20:2313–9.
62. Tjandrawinata RR, Dahiya R, Hughes-Fulford M. Induction of cyclo-oxygenase-2 mRNA by prostaglandin E2 in human prostatic carcinoma cells. *Br J Cancer* 1997;75:1111–8.
63. Uotila P, Valve E, Martikainen P, et al. Increased expression of cyclooxygenase-2 and nitric oxide synthase-2 in human prostate cancer. *Urol Res* 2001;29:23–8.
64. Yoshimura R, Sano H, Masuda C, et al. Expression of cyclooxygenase-2 in prostate carcinoma. *Cancer* 2000;89:589–96.
65. Buckman SY, Gresham A, Hale P, et al. COX-2 expression is induced by UVB exposure in human skin: implications for the development of skin cancer. *Carcinogenesis* 1998;19:723–9.
66. Higashi Y, Kanekura T, Kanzaki T. Enhanced expression of cyclooxygenase (COX)-2 in human skin epidermal cancer cells: evidence for growth suppression by inhibiting COX-2 expression. *Int J Cancer* 2000;86:667–71.
67. Muller-Decker K, Reinerth G, Krieg P, et al. Prostaglandin-H-synthase isozyme expression in normal and neoplastic human skin. *Int J Cancer* 1999;82:648–56.
68. Lim HY, Joo HJ, Choi JH, et al. Increased expression of cyclooxygenase-2 protein in human gastric carcinoma. *Clin Cancer Res* 2000;6:519–25.
69. Murata H, Kawano S, Tsuji S, et al. Cyclooxygenase-2 overexpression enhances lymphatic invasion and metastasis in human gastric carcinoma. *Am J Gastroenterol* 1999;94:451–5.
70. Ohno R, Yoshinaga K, Fujita T, et al. Depth of invasion parallels increased cyclooxygenase-2 levels in patients with gastric carcinoma. *Cancer* 2001;91:1876–81.
71. Ratnasinghe D, Tangrea JA, Roth MJ, et al. Expression of cyclooxygenase-2 in human adenocarcinomas of the gastric cardia and corpus. *Oncol Rep* 1999;6:965–8.
72. Ristimaki A, Honkanen N, Jankala H, et al. Expression of cyclooxygenase-2 in human gastric carcinoma. *Cancer Res* 1997; 57:1276–80.
73. Soydan AS, Gaffen JD, Weech PK, et al. Cytosolic phospholipase A2, cyclo-oxygenases and arachidonate in human stomach tumours. *Eur J Cancer* 1997;33:1508–12.
74. Uefuji K, Ichikura T, Mochizuki H, et al. Expression of cyclooxygenase-2 protein in gastric adenocarcinoma. *J Surg Oncol* 1998;69:168–72.
75. Uefuji K, Ichikura T, Mochizuki H. Cyclooxygenase-2 expression is related to prostaglandin biosynthesis and angiogenesis in human gastric cancer. *Clin Cancer Res* 2000;6:135–8.
76. Uefuji K, Ichikura T, Mochizuki H. Expression of cyclooxygenase-2 in human gastric adenomas and adenocarcinomas. *J Surg Oncol* 2001;76:26–30.
77. Yamamoto H, Itoh F, Fukushima H, et al. Overexpression of cyclooxygenase-2 protein is less frequent in gastric cancers with microsatellite instability. *Int J Cancer* 1999;84:400–3.
78. Oshima M, Dinchuk JE, Kargman SL, et al. Suppression of intestinal polyposis in Apc delta716 knockout mice by inhibition of cyclooxygenase 2 (COX-2). *Cell* 1996;87:803–9.
79. Liu CH, Chang SH, Narko K, et al. Overexpression of cyclooxygenase-2 is sufficient to induce tumorigenesis in transgenic mice. *J Biol Chem* 2001;276:18563–9.
80. Eling TE, Thompson DC, Foureman GL, et al. Prostaglandin H synthase and xenobiotic oxidation. *Annu Rev Pharmacol Toxicol* 1990;30:1–45.
81. Windmill KF, McKinnon RA, Zhu X, et al. The role of xenobiotic metabolizing enzymes in arylamine toxicity and carcinogenesis: functional and localization studies. *Mutat Res* 1997;376:153–60.
82. Tsujii M, Kawano S, Tsuji S, et al. Cyclooxygenase regulates angiogenesis induced by colon cancer cells. *Cell* 1998;93:705–16.
83. Hernandez GL, Volpert OV, Iniguez MA, et al. Selective inhibition of vascular endothelial growth factor-mediated angiogenesis by cyclosporin A: roles of the nuclear factor of activated T cells and cyclooxygenase 2. *J Exp Med* 2001;193:607–20.
84. Seed MP, Brown JR, Freemantle CN, et al. The inhibition of colon-26 adenocarcinoma development and angiogenesis by topical diclofenac in 2.5% hyaluronan. *Cancer Res* 1997;57:1625–9.
85. Tsujii M, DuBois RN. Alterations in cellular adhesion and apoptosis in epithelial cells overexpressing prostaglandin endoperoxide synthase 2. *Cell* 1995;83:493–501.
86. Zhang L, Yu J, Park BH, et al. Role of BAX in the apoptotic response to anticancer agents. *Science* 2000;290:989–92.
87. Li M, Lotan R, Levin B, et al. Aspirin induction of apoptosis in esophageal cancer: a potential for chemoprevention. *Cancer Epidemiol Biomarkers Prev* 2000;9:545–9.
88. Hsueh CT, Chiu CF, Kelsen DP, et al. Selective inhibition of cyclooxygenase-2 enhances mitomycin-C-induced apoptosis. *Cancer Chemother Pharmacol* 2000;45:389–96.
89. Grossman EM, Longo WE, Panesar N, et al. The role of cyclooxygenase enzymes in the growth of human gall bladder cancer cells. *Carcinogenesis* 2000;21:1403–9.
90. Petersen C, Petersen S, Milas L, et al. Enhancement of intrinsic tumor cell radiosensitivity induced by a selective cyclooxygenase-2 inhibitor. *Clin Cancer Res* 2000;6:2513–20.
91. Nishimura G, Yanoma S, Mizuno H, et al. A selective cyclooxygenase-2 inhibitor suppresses tumor growth in nude mouse xenografted with human head and neck squamous carcinoma cells. *Jpn J Cancer Res* 1999;90:1152–62.
92. Zhang G, Tu C, Zhou G, et al. Indomethacin induces apoptosis and inhibits proliferation in chronic myeloid leukemia cells. *Leuk Res* 2000;24:385–92.
93. Hida T, Kozaki K, Muramatsu H, et al. Cyclooxygenase-2 inhibitor induces apoptosis and enhances cytotoxicity of various anticancer agents in non-small cell lung cancer cell lines. *Clin Cancer Res* 2000;6:2006–11.
94. Liu XH, Yao S, Kirschenbaum A, et al. NS398, a selective cyclooxygenase-2 inhibitor, induces apoptosis and down-regulates bcl-2 expression in LNCaP cells. *Cancer Res* 1998;58:4245–9.
95. Gupta RA, DuBois RN. Translational studies on Cox-2 inhibitors in the prevention and treatment of colon cancer. *Ann NY Acad Sci* 2000;910:196–204; discussion 204–6.
96. Waddell WR. Stimulation of apoptosis by sulindac and piroxicam. *Clin Sci (Colch)* 1998;95:385–8.
97. Balch CM, Dougherty PA, Cloud GA, et al. Prostaglandin E2-mediated suppression of cellular immunity in colon cancer patients. *Surgery* 1984;95:71–7.
98. Weitzman SA, Gordon LI. Inflammation and cancer: role of phagocyte-generated oxidants in carcinogenesis. *Blood* 1990;76: 655–63.
99. Shreedhar V, Giese T, Sung VW, et al. A cytokine cascade including prostaglandin E2, IL-4, and IL-10 is responsible for UV-induced systemic immune suppression. *J Immunol* 1998;160: 3783–9.
100. Plescia OJ, Smith AH, Grinwich K. Subversion of immune system by tumor cells and role of prostaglandins. *Proc Natl Acad Sci U S A* 1975;72:1848–51.
101. Tsujii M, Kawano S, DuBois RN. Cyclooxygenase-2 expression in human colon cancer cells increases metastatic potential. *Proc Natl Acad Sci U S A* 1997;94:3336–40.
102. Masferrer JL, Leahy KM, Koki AT, et al. Antiangiogenic and antitumor activities of cyclooxygenase-2 inhibitors. *Cancer Res* 2000;60:1306–11.

103. Tomozawa S, Nagawa H, Tsuno N, et al. Inhibition of haematogenous metastasis of colon cancer in mice by a selective COX-2 inhibitor, JTE-522. *Br J Cancer* 1999;81:1274–9.
104. Jiang MC, Liao CF, Lee PH. Aspirin inhibits matrix metalloproteinase-2 activity, increases E-cadherin production, and inhibits in vitro invasion of tumor cells. *Biochem Biophys Res Commun* 2001;282:671–7.
105. Taketo MM. Cyclooxygenase-2 inhibitors in tumorigenesis (part I). *J Natl Cancer Inst* 1998;90:1529–36.
106. Jacoby RF, Seibert K, Cole CE, et al. The cyclooxygenase-2 inhibitor celecoxib is a potent preventive and therapeutic agent in the min mouse model of adenomatous polyposis. *Cancer Res* 2000;60:5040–4.
107. Reddy BS, Hirose Y, Lubet R, et al. Chemoprevention of colon cancer by specific cyclooxygenase-2 inhibitor, celecoxib, administered during different stages of carcinogenesis. *Cancer Res* 2000;60:293–7.
108. Oshima M, Murai N, Kargman S, et al. Chemoprevention of intestinal polyposis in the Apcdelta716 mouse by rofecoxib, a specific cyclooxygenase-2 inhibitor. *Cancer Res* 2001;61:1733–40.
109. Haedersdal M, Poulsen T, Wulf HC. Effects of systemic indomethacin on photocarcinogenesis in hairless mice. *J Cancer Res Clin Oncol* 1995;121:257–61.
110. Rubio CA. Further studies on the therapeutic effect of indomethacin on esophageal tumors. *Cancer* 1986;58:1029–31.
111. Castonguay A, Rioux N. Inhibition of lung tumourigenesis by sulindac: comparison of two experimental protocols. *Carcinogenesis* 1997;18:491–6.
112. Alshafie GA, Harris RE, Robertson FM, et al. Comparative chemopreventive activity of ibuprofen and N-(4-hydroxyphenyl) retinamide against the development and growth of rat mammary adenocarcinomas. *Anticancer Res* 1999;19:3031–6.
113. Wechter WJ, Leipold DD, Murray ED Jr, et al. E-7869 (R-flurbiprofen) inhibits progression of prostate cancer in the TRAMP mouse. *Cancer Res* 2000;60:2203–8.
114. Moon RC, Kelloff GJ, Detrisac CJ, et al. Chemoprevention of OH-BBN-induced bladder cancer in mice by piroxicam. *Carcinogenesis* 1993;14:1487–9.
115. Potter M. Indomethacin inhibition of pristane plasmacytomagenesis in genetically susceptible inbred mice. *Adv Exp Med Biol* 1999;469:151–6.
116. Potter M, Wax J, Jones GM. Indomethacin is a potent inhibitor of pristane and plastic disc induced plasmacytomagenesis in a hypersusceptible BALB/c congenic strain. *Blood* 1997;90:260–9.
117. Taketo MM. Cyclooxygenase-2 inhibitors in tumorigenesis (Part II). *J Natl Cancer Inst* 1998;90:1609–20.
118. Giovannucci E. The prevention of colorectal cancer by aspirin use. *Biomed Pharmacother* 1999;53:303–8.
119. Paganini-Hill A. Aspirin and colorectal cancer: the Leisure World cohort revisited. *Prev Med* 1995;24:113–5.
120. Sturmer T, Glynn RJ, Lee IM, et al. Aspirin use and colorectal cancer: post-trial follow-up data from the Physicians' Health Study. *Ann Intern Med* 1998;128:713–20.
121. Castelao JE, Yuan JM, Gago-Dominguez M, et al. Non-steroidal anti-inflammatory drugs and bladder cancer prevention. *Br J Cancer* 2000;82:1364–9.
122. Coogan PF, Rao SR, Rosenberg L, et al. The relationship of nonsteroidal anti-inflammatory drug use to the risk of breast cancer. *Prev Med* 1999;29:72–6.
123. Egan KM, Stampfer MJ, Giovannucci E, et al. Prospective study of regular aspirin use and the risk of breast cancer. *J Natl Cancer Inst* 1996;88:988–93.
124. Harris RE, Namboodiri KK, Farrar WB. Nonsteroidal antiinflammatory drugs and breast cancer. *Epidemiology* 1996;7:203–5.
125. Harris RE, Kasbari S, Farrar WB. Prospective study of nonsteroidal anti-inflammatory drugs and breast cancer. *Oncol Rep* 1999;6:71–3.
126. Schreinemachers DM, Everson RB. Aspirin use and lung, colon, and breast cancer incidence in a prospective study *Epidemiology* 1994;5:138–46.
127. Sharpe CR, Collet JP, McNutt M, et al. Nested case-control study of the effects of non-steroidal anti-inflammatory drugs on breast cancer risk and stage. *Br J Cancer* 2000;83:112–20.
128. Langman MJ, Cheng KK, Gilman EA, et al. Effect of anti-inflammatory drugs on overall risk of common cancer: case-control study in general practice research database. *BMJ* 2000; 320:1642–6.
129. Thun MJ, Namboodiri MM, Calle EE, et al. Aspirin use and risk of fatal cancer. *Cancer Res* 1993;53:1322–7.
130. Funkhouser EM, Sharp GB. Aspirin and reduced risk of esophageal carcinoma. *Cancer* 1995;76:1116–9.
131. Farrow DC, Vaughan TL, Hansten PD, et al. Use of aspirin and other nonsteroidal anti-inflammatory drugs and risk of esophageal and gastric cancer. *Cancer Epidemiol Biomarkers Prev* 1998;7:97–102.
132. Zaridze D, Borisova E, Maximovitch D, et al. Aspirin protects against gastric cancer: results of a case-control study from Moscow, Russia. *Int J Cancer* 1999;82:473–6.
133. Thun MJ, Heath CW Jr. Aspirin use and reduced risk of gastrointestinal tract cancers in the American Cancer Society prospective studies. *Prev Med* 1995;24:116–8.
134. Hawk E, Lubet R, Limburg P. Chemoprevention in hereditary colorectal cancer syndromes. *Cancer* 1999;86:2551–63.
135. Waddell WR, Loughry RW. Sulindac for polyposis of the colon. *J Surg Oncol* 1983;24:83–7.
136. Hixson LJ, Earnest DL, Fennerty MB, et al. NSAID effect on sporadic colon polyps *Am J Gastroenterol* 1993;88:1652–6.
137. Ladenheim J, Garcia G, Titzer D, et al. Effect of sulindac on sporadic colonic polyps *Gastroenterology* 1995;108:1083–7.
138. Matsuhashi N, Nakajima A, Fukushima Y, et al. Effects of sulindac on sporadic colorectal adenomatous polyps *Gut* 1997; 40:344–9.
139. Gann PH, Manson JE, Glynn RJ, et al. Low-dose aspirin and incidence of colorectal tumors in a randomized trial. *J Natl Cancer Inst* 1993;85:1220–4.
140. Brooks P. Use and benefits of nonsteroidal anti-inflammatory drugs. *Am J Med* 1998;104:9S-13S; discussion 21S–22S.
141. Estensen RD, Wattenberg LW. Studies of chemopreventive effects of myo-inositol on benzo[a]pyrene- induced neoplasia of the lung and forestomach of female A/J mice. *Carcinogenesis* 1993; 14:1975–7.
142. Wattenberg LW, Estensen RD. Chemopreventive effects of myo-inositol and dexamethasone on benzo[a]pyrene and 4-(methylnitrosoamino)-1-(3-pyridyl)-1-butanone-induced pulmonary carcinogenesis in female A/J mice. *Cancer Res* 1996;56:5132–5.
143. Wattenberg LW, Wiedmann TS, Estensen RD, et al. Chemoprevention of pulmonary carcinogenesis by aerosolized budesonide in female A/J mice. *Cancer Res* 1997;57:5489–92.
144. Peters DC, Foster RH. Diclofenac/hyaluronic acid. *Drugs Aging* 1999;14:313–9; discussion 320–1.
145. Ruffin MTT, Krishnan K, Rock CL, et al. Suppression of human colorectal mucosal prostaglandins: determining the lowest effective aspirin dose. *J Natl Cancer Inst* 1997;89:1152–60.
146. Calaluce R, Earnest DL, Heddens D, et al. Effects of piroxicam on prostaglandin E2 levels in rectal mucosa of adenomatous polyp patients: a randomized phase IIb trial. *Cancer Epidemiol Biomarkers Prev* 2000;9:1287–92.
147. Gupta RA, DuBois RN. Aspirin, NSAIDS, and colon cancer prevention: mechanisms? *Gastroenterology* 1998;114:1095–8.
148. Piazza GA, Alberts DS, Hixson LJ, et al. Sulindac sulfone inhibits azoxymethane-induced colon carcinogenesis in rats without reducing prostaglandin levels. *Cancer Res* 1997;57:2909–15.
149. Reddy BS, Kawamori T, Lubet RA, et al. Chemopreventive efficacy of sulindac sulfone against colon cancer depends on time of administration during carcinogenic process. *Cancer Res* 1999; 59:3387–91.
150. Mahmoud NN, Boolbol SK, Dannenberg AJ, et al. The sulfide metabolite of sulindac prevents tumors and restores enterocyte apoptosis in a murine model of familial adenomatous polyposis. *Carcinogenesis* 1998;19:87–91.
151. Wechter WJ, Murray ED Jr, Kantoci D, et al. Treatment and survival study in the C57BL/6J-APC(Min)/+(Min) mouse with R-flurbiprofen. *Life Sci* 2000;66:745–53.

152. van Stolk R, Stoner G, Hayton WL, et al. Phase I trial of exisulind (sulindac sulfone, FGN-1) as a chemopreventive agent in patients with familial adenomatous polyposis. *Clin Cancer Res* 2000;6:78–89.
153. McCracken JD, Wechter WJ, Liu Y, et al. Antiproliferative effects of the enantiomers of flurbiprofen. *J Clin Pharmacol* 1996;36:540–5.
154. Wechter WJ, Kantoci D, Murray ED Jr, et al. R-flurbiprofen chemoprevention and treatment of intestinal adenomas in the APC(Min)/+ mouse model: implications for prophylaxis and treatment of colon cancer. *Cancer Res* 1997;57:4316–24.
155. Silverstein FE, Faich G, Goldstein JL, et al. Gastrointestinal toxicity with celecoxib vs nonsteroidal anti-inflammatory drugs for osteoarthritis and rheumatoid arthritis: the CLASS study: a randomized controlled trial. Celecoxib Long-term Arthritis Safety Study. *JAMA* 2000;284:1247–55.
156. Vane JR, Botting RM. Mechanism of action of antiinflammatory drugs. *Int J Tissue React* 1998;20:3–15.
157. Takahashi M, Fukutake M, Yokota S, et al. Suppression of azoxymethane-induced aberrant crypt foci in rat colon by nimesulide, a selective inhibitor of cyclooxygenase 2. *J Cancer Res Clin Oncol* 1996;122:219–22.
158. Reddy BS, Rao CV, Seibert K. Evaluation of cyclooxygenase-2 inhibitor for potential chemopreventive properties in colon carcinogenesis. *Cancer Res* 1996;56:4566–9.
159. Fukutake M, Nakatsugi S, Isoi T, et al. Suppressive effects of nimesulide, a selective inhibitor of cyclooxygenase-2, on azoxymethane-induced colon carcinogenesis in mice. *Carcinogenesis* 1998;19:1939–42.
160. Kawamori T, Rao CV, Seibert K, et al. Chemopreventive activity of celecoxib, a specific cyclooxygenase-2 inhibitor, against colon carcinogenesis. *Cancer Res* 1998;58:409–12.
161. Yoshimi N, Kawabata K, Hara A, et al. Inhibitory effect of NS-398, a selective cyclooxygenase-2 inhibitor, on azoxymethane-induced aberrant crypt foci in colon carcinogenesis of F344 rats. *Jpn J Cancer Res* 1997;88:1044–51.
162. Yoshimi N, Shimizu M, Matsunaga K, et al. Chemopreventive effect of N-(2-cyclohexyloxy-4-nitrophenyl)methane sulfonamide (NS-398), a selective cyclooxygenase-2 inhibitor, in rat colon carcinogenesis induced by azoxymethane. *Jpn J Cancer Res* 1999;90:406–12.
163. Alshafie GA, Abou-Issa HM, Seibert K, et al. Chemotherapeutic evaluation of celecoxib, a cyclooxygenase-2 inhibitor, in a rat mammary tumor model. *Oncol Rep* 2000;7:1377–81.
164. Harris RE, Alshafie GA, Abou-Issa H, et al. Chemoprevention of breast cancer in rats by celecoxib, a cyclooxygenase 2 inhibitor. *Cancer Res* 2000;60:2101–3.
165. Matsunaga K, Yoshimi N, Yamada Y, et al. Inhibitory effects of nabumetone, a cyclooxygenase-2 inhibitor, and esculetin, a lipoxygenase inhibitor, on N-methyl-N-nitrosourea-induced mammary carcinogenesis in rats. *Jpn J Cancer Res* 1998;89:496–501.
166. Fischer SM, Lo HH, Gordon GB, et al. Chemopreventive activity of celecoxib, a specific cyclooxygenase-2 inhibitor, and indomethacin against ultraviolet light-induced skin carcinogenesis. *Mol Carcinog* 1999;25:231–40.
167. Pentland AP, Schoggins JW, Scott GA, et al. Reduction of UV-induced skin tumors in hairless mice by selective COX-2 inhibition. *Carcinogenesis* 1999;20:1939–44.
168. Grubbs CJ, Lubet RA, Koki AT, et al. Celecoxib inhibits N-butyl-N-(4-hydroxybutyl)-nitrosamine-induced urinary bladder cancers in male B6D2F1 mice and female Fischer-344 rats *Cancer Res* 2000;60:5599–602.
169. Okajima E, Denda A, Ozono S, et al. Chemopreventive effects of nimesulide, a selective cyclooxygenase-2 inhibitor, on the development of rat urinary bladder carcinomas initiated by N-butyl-N-(4-hydroxybutyl)nitrosamine. *Cancer Res* 1998;58:3028–31.
170. Rioux N, Castonguay A. Prevention of NNK-induced lung tumorigenesis in A/J mice by acetylsalicylic acid and NS-398. *Cancer Res* 1998;58:5354–60.
171. Dolara P, Caderni G, Tonelli F. Nimesulide, a selective anti-inflammatory cyclooxygenase-2 inhibitor, does not affect polyp number and mucosal proliferation in familial adenomatous polyposis. *Scand J Gastroenterol* 1999;34:1168.
172. Steinbach G, Lynch PM, Phillips RK, et al. The effect of celecoxib, a cyclooxygenase-2 inhibitor, in familial adenomatous polyposis. *N Engl J Med* 2000;342:1946–52.
173. Rostom A, Wells G, Tugwell P, et al. The prevention of chronic NSAID induced upper gastrointestinal toxicity: a Cochrane collaboration metaanalysis of randomized controlled trials. *J Rheumatol* 2000;27:2203–14.
174. Sporn MB. Combination chemoprevention of cancer. *Nature* 1980;287:107–8.
175. Reddy BS, Hirose Y, Cohen LA, et al. Preventive potential of wheat bran fractions against experimental colon carcinogenesis: implications for human colon cancer prevention. *Cancer Res* 2000;60:4792–7.
176. Nigro ND, Bull AW, Boyd ME. Inhibition of intestinal carcinogenesis in rats: effect of difluoromethylornithine with piroxicam or fish oil. *J Natl Cancer Inst* 1986;77:1309–13.
177. Rao CV, Tokumo K, Rigotty J, et al. Chemoprevention of colon carcinogenesis by dietary administration of piroxicam, alpha-difluoromethylornithine, 16 alpha-fluoro-5-androsten- 17-one, and ellagic acid individually and in combination. *Cancer Res* 1991;51:4528–34.
178. Reddy BS, Nayini J, Tokumo K, et al. Chemoprevention of colon carcinogenesis by concurrent administration of piroxicam, a nonsteroidal antiinflammatory drug with D,L-alpha-difluoromethylornithine, an ornithine decarboxylase inhibitor, in diet. *Cancer Res* 1990;50:2562–8.
179. Li H, Schut HA, Conran P, et al. Prevention by aspirin and its combination with alpha-difluoromethylornithine of azoxymethane-induced tumors, aberrant crypt foci and prostaglandin E2 levels in rat colon. *Carcinogenesis* 1999;20:425–30.
180. Jacoby RF, Cole CE, Tutsch K, et al. Chemopreventive efficacy of combined piroxicam and difluoromethylornithine treatment of Apc mutant Min mouse adenomas, and selective toxicity against Apc mutant embryos. *Cancer Res* 2000;60:1864–70.
181. Agarwal B, Rao CV, Bhendwal S, et al. Lovastatin augments sulindac-induced apoptosis in colon cancer cells and potentiates chemopreventive effects of sulindac. *Gastroenterology* 1999;117:838–47.
182. Torrance CJ, Jackson PE, Montgomery E, et al. Combinatorial chemoprevention of intestinal neoplasia *Nat Med* 2000;6:1024–8.
183. Mann M, Sheng H, Shao J, et al. Targeting cyclooxygenase 2 and HER-2/neu pathways inhibits colorectal carcinoma growth. *Gastroenterology* 2001;120:1713–9.
184. Lala PK, Elkashab M, Kerbel RS, et al. Cure of human melanoma lung metastases in nude mice with chronic indomethacin therapy combined with multiple rounds of IL-2: characteristics of killer cells generated in situ. *Int Immunol* 1990;2:1149–58.
185. Sawaoka H, Kawano S, Tsuji S, et al. Cyclooxygenase-2 inhibitors suppress the growth of gastric cancer xenografts via induction of apoptosis in nude mice. *Am J Physiol* 1998;274:G1061–7.
186. Stratton JA, Braly PS, DiSaia PJ. Response of human adenocarcinoma to chemotherapy: as sole agents and in combination with sodium ibuprofen. *Gynecol Oncol* 1984;18:145–9.
187. Heneghan JB. Inhibition of human colon tumor growth in nude mice by indomethacin. *Prog Clin Biol Res* 1985;181:315–8.
188. Goldman AP, Williams CS, Sheng H, et al. Meloxicam inhibits the growth of colorectal cancer cells. *Carcinogenesis* 1998;19:2195–9.
189. Goluboff ET, Shabsigh A, Saidi JA, et al. Exisulind (sulindac sulfone) suppresses growth of human prostate cancer in a nude mouse xenograft model by increasing apoptosis. *Urology* 1999;53:440–5.
190. Lundholm K, Gelin J, Hyltander A, et al. Anti-inflammatory treatment may prolong survival in undernourished patients with metastatic solid tumors. *Cancer Res* 1994;54:5602–6.
191. Sandler RS. Aspirin and other nonsteroidal anti-inflammatory agents in the prevention of colorectal cancer. *Important Adv Oncol* 1996;123–37.
192. Pascucci RA. COX-2-specific inhibition: implications for clinical practice. *J Am Osteopath Assoc* 1999;99:S18–22.
193. Weaver AL. Rofecoxib: clinical pharmacology and clinical experience. *Clin Ther* 2001;23:1323–38.