

## Coxibs and Other Nonsteroidal Anti-Inflammatory Drugs in Animal Models of Cancer Chemoprevention

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### Abstract

Coxibs, including celecoxib, and other nonsteroidal anti-inflammatory drugs (NSAID), including aspirin, are among the most promising cancer chemopreventive agents in development today. This article examines the data on the efficacy of these agents in animal model studies of cancer prevention carried out by the authors. The studies evaluated here are restricted to our rodent models of colon/intestinal, bladder, and nonmelanoma skin cancer, in which celecoxib and other NSAIDs were administered as either cancer preventive or therapeutic agents. These studies may shed light on several questions. Is celecoxib unique compared with other NSAIDs, and if so, what implications would this have for human use? Are standard NSAIDs (which inhibit both COX-1 and COX-2) as effective as celecoxib in animal studies? Is the efficacy of celecoxib in particular or NSAIDs in general due to their off-target effects or to their effects on COX-1 and COX-2? What is the likely efficacy of low-dose aspirin? Some questions raised by human trials and epidemiology are discussed and related to our observations in animal model studies. We also discuss the problem of cardiovascular (CV) events associated with coxibs and certain other NSAIDs and whether results in animal models are predictive of efficacy in humans. On the basis of epidemiologic studies and its CV profile, aspirin seems to be the most promising NSAID for preventing human colorectal, bladder, and skin cancer, although the animal data for aspirin are less clear. A comprehensive understanding of the results of coxibs and other NSAIDs in animal studies may help inform and shape human trials of these commonly employed, relatively inexpensive, and highly effective compounds. *Cancer Prev Res*; 4(11); 1728–35. ©2011 AACR.

### Introduction

This minireview is based primarily upon animal model studies done by the authors that we feel can shed light on questions about the use of COX-2 inhibitors (coxibs) and other nonsteroidal anti-inflammatory drugs (NSAID) in the chemoprevention of cancer in humans. The minireview is not all-inclusive and examines results only in models of colon/intestine, urinary bladder, and squamous cell cancer of the skin. Nevertheless, the studies presented here contribute data toward answering certain generalized questions including the following: Is COX-2 the primary prevention target of coxibs and other NSAIDs? Is there something unique about celecoxib as contrasted with other NSAIDs? Are NSAIDs/coxibs effective when taken later in tumor

progression? Can one achieve a safe and effective NSAID/coxib regimen for human use? We address these questions directly in the final section.

### Prostaglandin Synthesis: Multiple Levels of Regulation

Prostaglandins (PG) are one of the most abundant members of the eicosanoid family of arachidonic acid-derived autacoids. Arachidonic acid, a 20-carbon, 4-double bond fatty acid, is either obtained from the diet or, to a much lesser extent, is synthesized from linoleic acid, an 18-carbon, 2-double bond dietary fatty acid. Arachidonic acid is normally stored esterified to the glycerol backbone of membrane phospholipids; in this form, it cannot be metabolized to PGs. Following hydrolysis by phospholipase A<sub>2</sub>, which is one of the rate-limiting steps for PG synthesis, arachidonic acid is available as a substrate for enzymatic oxidation by several different enzyme systems including COXs, lipoxygenases, and cytochrome P450s. PG synthesis is thus regulated at several levels including substrate abundance and availability, the level of COX expression (including the 2 isoforms, COX-1 and COX-2), and the level of expression of the PG synthases. COX-1 is expressed constitutively, whereas COX-2, which is nearly undetectable in most epithelial tissues under normal conditions, is highly

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upregulated by a wide variety of physical irritants, growth factors, cytokines, etcetera. The biological activities, particularly those related to inflammation, of the 3 major PG products of COX-1 and COX-2, namely PGE<sub>2</sub>, PGF<sub>2α</sub>, and PGD<sub>2</sub>, were not understood until Vane and colleagues showed that aspirin inhibited PG synthesis as well as inflammation (1, 2). Other nonselective COX-inhibiting drugs, for example, indomethacin, that inhibit PG synthesis were subsequently developed and collectively named NSAIDs. These earlier NSAIDs are a heterogeneous group of compounds that are often chemically unrelated but have similar mechanisms of action and typically similar side effects, including gastrointestinal (GI) complications (Fig. 1).

The recognition that there are 2 isoforms of COX suggested that they have different biological activities (2). The PGs derived from COX-1 are responsible for homeostatic maintenance of the GI mucosa and for smooth muscle contraction. COX-2, on the other hand, is induced during inflammation, including arthritis, and is overexpressed in many epithelial tumors. These observations led pharmaceutical companies to develop selective COX-2-inhibiting NSAIDs referred to as coxibs, which reduced inflammation with a decreased propensity for GI complications. On the basis of observed upregulation of COX-2 in many cancers, the chemopreventive activity of coxibs was examined in animal models.

### Colorectal Cancer

The hypothesis that NSAIDs might prevent the occurrence or severity of colorectal cancer (CRC) arose from studies showing that PGE<sub>2</sub> levels were higher in CRC than surrounding normal tissue (3–5). In addition, many epidemiologic studies showed that long-term use of aspirin and other NSAIDs was associated with a significant reduction in risk of CRC (3, 6), including a very recent prospective study on more than 300,000 men and women that showed a significant reduction in risk of CRC with aspirin and nonaspirin NSAIDs (7). The 2 models of GI neoplasia most commonly used in identifying preventive activity are the rodent azoxymethane (AOM) carcinogen model and the genetically modified Min mouse model. The adenomatous polyps and carcinomas induced by AOM are similar to those observed in humans (8). AOM induces minimally invasive colon cancer in rats and mice, which have mutations in the Wnt pathway, for example, adenomatous polyposis coli (APC), beta-catenin. The Min mouse has a germline mutation in the APC gene and primarily develops spontaneous small intestinal neoplasias (9).

The first uses of NSAIDs in models of colon cancer showed that indomethacin was highly effective when treatment was initiated prior to AOM treatment (10). These initial studies led to a series of studies with enolic acids (e.g., piroxicam) and the propionic acid derivatives (e.g., ibuprofen; ref. 11). In most early studies, treatment was initiated when no carcinogen-initiated cells were present, suggesting that indomethacin, piroxicam, or other NSAIDs may either

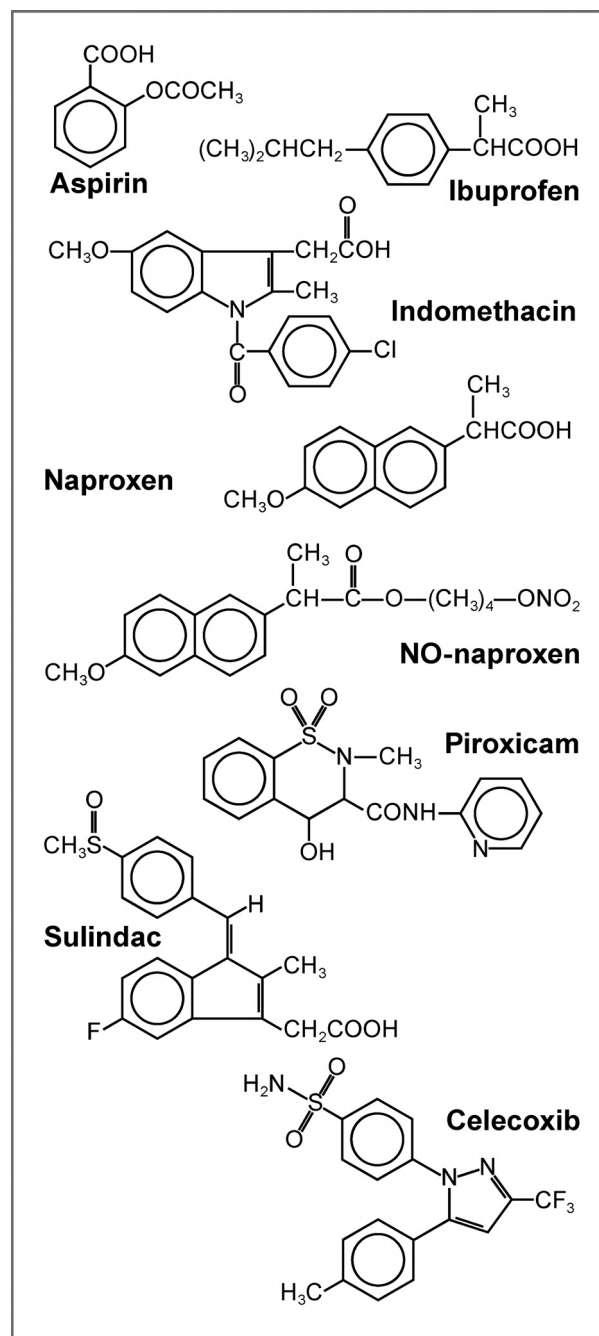


Figure 1. Chemical structure of common NSAIDs and of the selective COX-2-inhibiting NSAID celecoxib.

inhibit activation of AOM or inhibit the earliest stages of carcinogenesis. Many of these NSAIDs decreased colon tumor multiplicity by 65% to 80% in the AOM model (10, 11), which showed that NSAIDs of varied structures (Fig. 1) were effective in preventing colon cancer. We have recently undertaken studies with naproxen, a 2-arylpropionic acid derivative, which seems to be associated with minimal cardiovascular (CV) effects (12). This agent is highly effective in preventing both aberrant crypt foci (ACF)

and colon tumors at various doses (13). Subsequent to the studies showing NSAIDs were effective when initiated early, it was shown that piroxicam was also highly effective when initiated later in tumor progression when ACF and even adenomas already existed (14, 15).

The first study using dietary celecoxib found that a dose of 1,500 ppm decreased CRC multiplicity by 95% in the AOM rat model, which seemed more effective than most NSAIDs (16). A second study testing doses of 500, 1,000, and 1,500 ppm in the diet showed a dose-dependent decrease in colon cancer incidence (55%, 62%, and 77% reduction, respectively), and multiplicity (67%, 73%, and 84% reduction, respectively; ref. 17). In addition, we found that administering celecoxib starting at 14 weeks after the second AOM treatment was still able to reduce incidence and multiplicity by approximately 60% (17), which confirmed prior findings with piroxicam. These studies (Table 1) suggested that celecoxib could be an effective preventive agent for the secondary prevention of colon cancer in patients with familial adenomatous polyposis (FAP) or sporadic polyps. In the Adenoma Prevention with Celecoxib (APC) trial, celecoxib at doses of 400 mg twice daily significantly reduced the development of advanced adenomas by 55% to 65% (18), although this efficacy was achieved at doses that increased serious adverse CV events by 2- to 3-fold. In the PreSAP trial, celecoxib administered at 400 mg once a

day significantly reduced advanced adenomas by roughly 50%, without increasing CV events (19). A subsequent pooled analysis of safety data arising from 6 placebo-controlled trials showed that celecoxib was associated with an increased risk of serious adverse CV events, but that these risks were strongly associated with baseline CV risk, suggesting there may be individuals who could be treated safely (20). Furthermore, results of a recent analysis from the APC trial suggest that a low baseline level of the inflammatory and CV disease marker C-reactive protein is not associated with an increased risk of serious adverse CV effects in people taking celecoxib, even at the high dose of 400 mg twice daily (21). However, the hypothesis that low baseline CV risk or C-reactive protein correlates with the CV safety of celecoxib needs to be prospectively tested to be confirmed.

Even though aspirin is one of the most commonly consumed NSAIDs by humans, few prevention studies have been carried out in animal models of colon cancer. Our own studies in rats indicated that a relatively high dose of aspirin (1,800 ppm) was required to achieve a significant decrease in colon tumors (22). In contrast, efficacy was achieved at a dose of 600 ppm for prevention of ACF. There have been some reports of efficacy with aspirin at lower doses (400 ppm), but typically, those results show somewhat lower effectiveness and are still substantially above the dose equivalent of low-dose aspirin in humans based on normal

**Table 1.** Effects of NSAIDs or celecoxib in animal models

Model	Species	NSAID (ppm)	Relative Efficacy (%↓)	HED	Ref.
AOM colon	rats	Naproxen (400 ppm)	++ (60%)	320 mg	13
AOM colon	rats	Aspirin (200 ppm)	+ (20%)	160 mg	21
AOM colon	rats	Aspirin (1,800 ppm)	++ (60%)	1,440 mg	21
AOM colon	rats	Celecoxib (1,500 ppm)	++++ (96%)	1,200 mg	16
AOM colon	rats	Celecoxib (1,500 ppm)	+++ (84%)	1,200 mg	17
AOM colon	rats	Celecoxib (500 ppm)	++ (67%)	400 mg	17
Min intestine	mice	Aspirin (250 ppm)	++ (55%)	265 mg	31
Min intestine	mice	Celecoxib (1,500 ppm)	+++ (70%)	1,590 mg	28
UV skin	mice	Celecoxib (500 ppm)	+++ (70%)	533 mg	39
UV skin	mice	Celecoxib (150 ppm)	++ (60%)	160 mg	39
UV skin	mice	Indomethacin (4 ppm)	+++ (70%)	4.3 mg	39
UV skin	mice	Naproxen (400 ppm)	+++ (70)	427 mg	Unpublished data
OHBBN bladder	rats	Celecoxib (1,000 ppm)	++++ (90%)	800 mg	36
OHBBN bladder	mice	Celecoxib (1,250 ppm)	++++ (90%)	1,333 mg	36
OHBBN bladder	rats	Naproxen (400 ppm)	++++ (87%)	320 mg	35
OHBBN bladder	rats	Aspirin (300 ppm)	0	240 mg	35
OHBBN bladder	rats	Aspirin (3,000 ppm)	++ (65%)	2,400 mg	35

NOTE: The calculations below are standard scaling factors that would be used for the FDA. They do not take into account specific pharmacokinetics of individual agents which can only properly be done after gavage dosing.

HEDs were calculated as follows, using 100 ppm (100 µg/g diet) as an example. Rats, which eat 15 g food daily, would consume 1.5 mg drug; for a 250 g rat, the daily weight-based dose would be 6 mg drug/kg body weight. Dividing by the rat-to-human scaling factor of 6, the HED is 1 mg/kg body weight; for an 80 kg human this is 80 mg. Mice, which eat 4 g food daily, would consume 0.4 mg drug; for a 25 g mouse, the daily weight-based dose would be 16 mg drug/kg body weight. Dividing by the mouse-to-human scaling factor of 12, the HED is 1.33 mg/kg body weight; for an 80 kg human this is 106 mg.

Abbreviation: HED, human equivalent dose.

U.S. Food and Drug Administration (FDA) scaling factors (23).

### Intestinal Tumors in the Min Mouse

FAP is an autosomal dominantly inherited syndrome characterized by the development of multiple colorectal adenomas, some of which progress to malignancy. Groden and colleagues (24) were among the first to show that the disease is due to heritable mutations in the *APC* gene. *APC* mutations have also been shown to be common in sporadic colon cancers as well (25). The Min mouse, which carries a mutation in the *APC* gene, mimics the rapid development of numerous polyps that affect individuals with FAP (9). In an early study, piroxicam (200 ppm) significantly reduced tumor multiplicity in the Min model (26). Furthermore, sulindac, which inhibits polyp development in FAP patients (27) significantly decreased average tumor load in Min mice, further validating this model (28). When started early, celecoxib (1,500 ppm) reduced tumor multiplicity and tumor load by 70% and 83%, respectively, in the Min model, whereas starting treatment late, after most adenomas were established, still reduced tumor multiplicity and size by approximately 50% (29). Based in part on these preclinical studies, a double-blind, placebo-controlled clinical trial with celecoxib in FAP patients was done. After 6 months, patients receiving 400 mg twice daily had a 28% reduction in the number of colorectal polyps, whereas the 100 mg twice daily group showed an approximately 12% reduction. Although significant, and the basis for an FDA approval for celecoxib as an adjunct to usual standard of care in FAP patients, these results were not as dramatic as those from the animal studies. However, in the animal model the endpoint has routinely been preventing the development of new polyps, that is, tumor multiplicity, whereas in the human FAP trial the primary endpoint was polyp regression, although the endpoint included assessment of the overall polyp burden in an area, expressed as the sum of the diameter of the polyps, which includes both regression of existing polyps and prevention of new polyps in a defined area of the colon (30).

Several studies have been carried out in the Min model with aspirin (31). A review of these studies showed that a small reduction was seen in tumors of the small intestine, but no effect was observed on colonic polyps. In one early study, however, Min mice fed 250 or 500 ppm aspirin had an approximately 55% reduction in the number of intestinal tumors (32). Recently reported data suggested a trend toward a benefit of aspirin in a human phase III clinical trial in FAP patients (33). In other human clinical trials, aspirin consistently reduced sporadic colonic adenoma recurrence by 15% to 25% (34). However, this is markedly lower than that obtained in either celecoxib (40%–45% reduction; ref. 18) or the combined sulindac-difluoromethylornithine (DFMO) trials (70% reduction; ref. 35); it is recognized, however, that comparing the efficacy of different agents used in unrelated trials can be misleading.

### Bladder Tumors

In both rats and mice *N*-butyl-*N*-(4-hydroxybutyl)-nitrosamine (OH-BBN) induces invasive urinary bladder cancers that are histologically similar to human transitional cell carcinomas. Early studies showed that ketoprofen and sulindac have strong preventive activity when administered beginning 1 week before 8 to 10 weeks of treatment with OH-BBN (36). Studies with aspirin have been more problematic with a number of groups showing minimal efficacy at an approximately 300 ppm dose but substantial activity at 3,000 ppm (37). On the other hand, celecoxib at doses of 500 or 1,000 ppm reduced palpable tumors by more than 80% when initiated after OH-BBN (38). We also found that celecoxib decreased bladder cancer in rats when treatment was initiated up to 3 months after the last dose of OH-BBN (37). Recently, we have explored the use of naproxen, which has the least CV problems of any of the traditional NSAIDs and nitric oxide (NO)-naproxen, based on the finding that adding a NO group to NSAIDs (Fig. 1) may help alleviate GI tract toxicity. At doses significantly below the standard human dose, naproxen and NO-naproxen reduced the development of large urinary tract tumors by approximately 80% (13), comparable with the effects of high-dose celecoxib (38). These agents were also highly effective in preventing the development of large palpable tumors when treatment was initiated after microscopic carcinomas of the bladder already existed (37).

A recently published presurgical study of celecoxib (400 mg twice daily) given for at least 14 days to 13 patients with invasive transitional cell carcinoma of the bladder showed a lack of residual cancer at resection in 3 patients and induction of apoptosis in an additional 7 patients (39). A recently reported randomized controlled trial of celecoxib (200 mg twice daily) showed a trend favoring celecoxib (vs. placebo) of an increased recurrence-free survival rate (40). These data suggest the efficacy of celecoxib/NSAIDs for bladder cancer intervention and justify further investigation, such as that being conducted in the United Kingdom in the phase III Bladder COX Inhibition Trial (BOXIT), which is planned to complete accrual in early 2012.

### Nonmelanoma Skin Cancer

Nonmelanoma skin cancer (NMSC) in mice can be elicited with either the classical 2-stage initiation–promotion protocol or by repetitive exposure to UV light. Both models induce a marked inflammatory response. This observation triggered the earliest study on the ability of NSAIDs to prevent NMSC, that is, topical indomethacin reduced skin tumor development by approximately 30% in the initiation–promotion model (41). More recently, we found that mice fed 150 or 500 ppm celecoxib showed a dose-dependent reduction (60% and 89%, respectively) in tumor multiplicity in the UV carcinogenesis model. Indomethacin (4 ppm) reduced tumor multiplicity by 78% (42), suggesting that celecoxib is not

markedly more effective. Subsequently, we found that celecoxib was relatively effective in causing the regression of preexisting skin cancers in the UV model (43). We also found that piroxicam and naproxen are highly effective in preventing UV-induced skin cancer (unpublished data). We have also shown that the NSAIDs and celecoxib that are effective in preventing NMSC also inhibit PGE<sub>2</sub> production in UV-exposed epidermis (ref. 42 and unpublished data). Thus, there is a strong correlation between short-term inhibition of PGE<sub>2</sub> and long-term efficacy in preventing NMSC.

To further examine whether the inhibition of COX-2 was responsible for the dramatic reduction in skin tumors seen in the UV studies, we took a genetic approach. Although the loss of 1 allele of *COX-1* had no effect on skin tumor development, the loss of only 1 allele of *COX-2* significantly reduced tumor development in response to UV exposure (44). In humans, topical diclofenac, a FDA approved NSAID with selectivity for COX-2 inhibition, is efficacious in treating actinic keratoses (45). A recent clinical trial showed that celecoxib significantly reduced the development of NMSC in individuals with actinic keratoses (46). Collectively, these studies strongly suggest that COX-2 is a critical target for preventing skin cancer in humans as well as mice.

## Controversies and Conclusions

Here we will address certain controversies that have arisen in the field and offer our perspective and speculation based in large part, but not exclusively, on *in vivo* preclinical studies of NSAIDs in carcinogenesis, in which we have been directly involved.

### Is there something unique about celecoxib?

One of the questions raised is whether celecoxib has some unique prevention efficacy as compared with most NSAIDs. The first study achieved a 95% reduction in colon cancer in the AOM model (16) in contrast to traditional NSAIDs, which reduced colon cancer by 70% to 80% (11, 13). However, in a subsequent study a similar dose achieved roughly an 85% effect similar to traditional NSAIDs (17) and the dose used, 1,500 ppm, is in excess of the standard human dose (250 ppm), based on scaling factors. A lower dose of celecoxib is somewhat effective in colon, bladder, and skin models, but no more effective than a wide variety of NSAIDs at their own human equivalent doses (Table 1). In humans, celecoxib at 400 mg twice daily (18) was more effective at polyp prevention than aspirin (34), but less effective than the combination of sulindac and DFMO (35). However, data in FAP patients implies that celecoxib even at these supraoptimal doses is no more effective than even a relatively low dose of sulindac alone (27). It is recognized, however, that the comparisons between celecoxib and NSAIDs needs to also take into consideration that there is variability in outcome between studies. Side-by-side comparisons of celecoxib and NSAIDs are needed to address this issue.

### Is COX-2 the primary target of NSAIDs/coxibs, and are there substantial off-target effects of NSAIDs/coxibs?

Another issue that has been raised is whether the preventive effects of the NSAIDs or coxibs are really due to inhibition of PG synthesis or are due, at least in part, to off-target effects. Celecoxib was observed to inhibit Akt activation, and this COX-2-independent activity is associated with its apoptotic activity in some cell types (47). The off-target effects of aspirin also seem to be cell type dependent. For example, aspirin induces apoptosis in cervical cancer cells through reduction of ErbB2 expression (48). In the case of sulindac, it produces a major metabolite (sulindac sulfone) with substantially different properties and targets (it does not inhibit COX-2). However, there are significant caveats. These off-target effects are routinely seen in cell culture, for example, celecoxib more than 25 μmol/L, at concentrations far higher than are achieved clinically in serum (4–5 μmol/L after 400 mg twice daily oral dosing; ref. 49). In the case of sulindac, although sulindac sulfone is not a COX-2 inhibitor, the other major metabolite, sulindac sulfide, is.

As shown in Fig. 1 and Table 1, a wide variety of NSAIDs and celecoxib are highly effective at preventing colon, bladder, and skin tumors. It is hard to imagine that such a structurally varied group of agents can have similar off-target effects, although it is possible that NSAID-specific off-target effects may contribute to the preventive action of any particular NSAID. Furthermore, the finding that knocking out the *COX-2* gene can inhibit tumor formation in colon and skin would be compatible with PG production being the primary target. In addition, Chan and colleagues (1) recently reported that aspirin reduced the risk of developing COX-2-expressing, but not other, CRCs. Given these results, the COX-2 hypothesis would seem to be consistent with the widest variety of data. In addition, the data in the UV-induced skin model showing that local inhibition of COX-2-induced PGE<sub>2</sub> is predictive of preventive efficacy is similarly compatible with a COX-2 target. If indeed inhibition of PG production is the primary target, then a nonspecific NSAID that would inhibit PG production by COX-1 and COX-2 should be effective because most tumors express COX-1 as well as COX-2.

### What about aspirin?

Aspirin, a salicylate, is the most commonly consumed NSAID. Multiple population-based case-control studies and several randomized, controlled trials show a significantly reduced risk of CRC in regular aspirin users (50), although efficacy depends on the dose and duration of exposure. For example, alternate day use of low-dose aspirin was reported to be ineffective in reducing the risk of CRC in the Women's Health Study (51) as well as in the Physicians' Health Study, although the study was terminated after 5 years (52). A recent compilation of data, however, showed that doses as low as 75 mg/d are effective in reducing CRC risk after extended dosing (53). The long duration of aspirin use required to prevent CRC may reflect the time required for cancer to develop from precursor lesions. By contrast,

animal data imply that high doses of aspirin are required for efficacy, which may be based on interspecies differences in metabolism. However, the finding that celecoxib was equally effective in inhibiting adenoma formation in individuals taking low doses of aspirin, as in those not taking aspirin, argues that low-dose aspirin is unlikely to be highly effective (18). These controversies make aspirin dosing decisions quite difficult and are further complicated because many potential participants in trials may already be taking low-dose aspirin for prevention of CV events. This positive attribute of low-dose aspirin was summarized in a recent meta-analysis of 9 randomized trials that concluded that aspirin decreased the risk for CV events and nonfatal myocardial infarctions (54). However, the effects of lower doses of aspirin required extended exposures often of 10 years or greater.

#### **How late in tumor progression can you wait?**

One might imagine that NSAIDs and coxibs, which were first proposed for use in a preventive setting, might be effective only in early cancer development. Thus, some of the epidemiologic data with colon cancer implied that striking efficacy was observed only after extended exposure of more than 10 years (55), which would be most compatible with efficacy early in tumor development. Furthermore, early animal studies support this view because most studies administered agents early in tumor development and continually. However, other studies showed that these agents were effective in later stages of colon cancer development when adenomas already existed (14, 15). Similarly, we found in skin and bladder that NSAIDs and coxibs are effective even when tumors are present (37, 42). This greater efficacy at later stages is consistent with the finding in various adenoma studies that NSAIDs seem more effective in prevention of advanced adenomas as contrasted with earlier adenomas. Human data on celecoxib effects on early-stage ACF are equivocal (56), and sulindac was found to be ineffective in reducing the number of ACF in patients with multiple/advanced colorectal adenomas (57). In addition, recent data in skin shows that although celecoxib was ineffective in blocking the formation of early-stage actinic keratosis, it reduced the formation of NMSC by 50% (46), arguing that NSAIDs/coxibs work further along in tumor progression. Finally, recent epidemiologic studies find that NSAIDs are effective even in patients being treated with standard therapies for advanced colon and breast cancers (1, 58). In another study, regular aspirin use after diagnosis of CRC reduced the risk of both CRC-specific and overall mortality (59).

#### **Can animal models predict the efficacy of NSAIDs in human trials?**

We have discussed the effects of NSAIDs/coxibs in colon/intestine, nonmelanoma skin, and bladder cancer. Multiple clinical efficacy trials employing NSAIDs/coxibs for blocking the development of sporadic colon adenomas have shown that these agents are effective, with aspirin showing more

limited activity than celecoxib or sulindac plus DFMO (18, 19, 35, 50). However, the relative contribution of sulindac and DFMO are not known, and there are caveats when comparing trials using different agents. Recent data indicating that aspirin was efficacious in a phase III trial for CRC prevention in patients with Lynch syndrome are consistent with recent positive aspirin data in a mouse model of Lynch syndrome (60). Recently, a clinical trial of oral celecoxib showed that this agent could inhibit the formation of squamous and basal cell skin cancers by roughly 60% (46). Furthermore, clinical data show that topical application of diclofenac is partially effective in preventing actinic keratoses and may be more effective in blocking SCCs of the skin (45). Thus, the clinical results seem to be in line with the high efficacy of late intervention observed in animal models.

#### **Can we identify a (relatively) nontoxic NSAID?**

Given the striking animal data, significant epidemiologic data in the colorectum and esophagus, and clinical trials, particularly, in CRC prevention (but more recently in skin cancer), the identification of an NSAID/coxib that can be used safely in a prevention setting is a high priority. The initial concerns with regard to NSAIDs were ulcers and potentially life-threatening bleeding. Although the incidence of these events is probably less than 1/10,000 for most NSAID users, they are serious concerns for individuals who might consider NSAIDs for CRC prevention, given the rarity of the disease in the general population as well as competing preventive strategies, such as endoscopic polypectomy. It seems that the COX-2 inhibitors are associated with significantly less upper GI toxicity (61). Thus, there was no significant increase in GI toxicity in the celecoxib trial (18, 19). However, rofecoxib and valdecoxib (at standard doses) seem to increase CV events. Although celecoxib at the standard dose does not significantly increase CV events, the higher doses used in adenoma prevention trials did. This led to an examination of most NSAIDs, and agents such as diclofenac clearly increase CV events and has led to a black-box warning. One NSAID that has consistently proven to have minimal CV effects, and potentially even to be cardioprotective, has been naproxen (12), although the data from the Alzheimer's Disease Anti-inflammatory Prevention Trial (ADAPT) is suggestive of increased CV risk (62). In addition, low-dose aspirin, as mentioned above, is both cardioprotective and chemopreventive when taken over an extended duration (18, 19). This raises the question of whether it is "better" to take (i) low-dose celecoxib, which probably has low gastric toxicity and limited CV effects but for which the prevention data are not as clear, or (ii) aspirin over an extended time period, which has a cardioprotective effect and for which there is the strongest epidemiologic evidence of prevention; however, questions arise with regard to an effective dose, or (iii) naproxen, which seems to have a good CV profile along with the potential use of a proton pump inhibitor to decrease GI events. At the moment, aspirin would seem to be the choice with the most epidemiologic evidence behind it. As with all prevention studies, the real question is the risk to benefit ratio and,

concomitantly, whether one can identify predictive markers of greater benefit (63) and lesser potential harm (64) from particular NSAIDs and whether one can define a "high-risk" group who has more to gain than lose through interventions of this nature.

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## Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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# Cancer Prevention Research

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