Celecoxib Colorectal Bioavailability and Chemopreventive Response in Patients with Familial Adenomatous Polyposis

Peiying Yang[1], Xiangsheng Zuo[2,3], Shailesh Advani[4], Bo Wei[1], Jessica Malek[2], Rena Sue Day[5], and Imad Shureiqi[2,3]

ABSTRACT

Why celecoxib exerts chemopreventive activity in only some familial adenomatous polyposis (FAP) patients remains poorly understood. We conducted a phase II clinical study to identify potential predictive biomarkers for celecoxib chemopreventive activity in FAP. Twenty-seven patients with FAP completed a 6-month oral course of 400 mg of celecoxib twice a day; they underwent colonoscopies before and after celecoxib treatment to assess colorectal polyp tumor burden and to obtain normal and polyp colorectal biopsies to measure celecoxib, 13-*S*-hydroxyoctadecadienoic acid (13-HODE), 15-HETE, 12-HETE, and LTB4 levels by LC/MS-MS. Celecoxib levels in sera from those patients were also measured before treatment and after 2, 4, and 6 months of treatment. Nineteen of the 27 patients experienced a response to celecoxib, with a ≥ 28% reduction of colonic polyp burden on the basis of a reproducible quantitative assessment of colonoscopy results. Celecoxib levels were significantly lower in polyp tissues than in normal colorectal tissues. Celecoxib levels in sera and normal colorectal tissues were correlated in patients who experienced a response to celecoxib but not in those who did not. Among the measured lipoxygenase products, only 13-HODE levels were significantly lower in polyp tissues than in normal tissues. Our findings demonstrate the differential bioavailability of celecoxib between normal and polyp tissues and its potential effects on clinical response in patients with FAP.

Prevention Relevance: This study evaluated potential predictive biomarkers for celecoxib chemopreventive activity in patients with FAP. Our findings demonstrated the differential bioavailability of celecoxib between normal and polyp tissues and its potential effects on clinical chemopreventive response in patients with FAP.

See related Spotlight, p. 205

Introduction

Non-steroidal anti-inflammatory drugs (NSAID) are a very promising class of chemopreventive agents in colorectal carcinogenesis, as shown in epidemiologic, preclinical, and clinical studies (1). Nevertheless, the estimated efficacy rate of NSAIDs in colorectal carcinogenesis chemoprevention is 50% or less (1–4). Furthermore, NSAIDs, especially COX-2 inhibitors such as celecoxib, have been associated with an increased risk of cardiovascular events (5, 6). Thus, identifying patients who might benefit from this chemopreventive approach is imperative to better tailor treatment to maximize the benefit versus the risk.

Familial adenomatous polyposis (FAP) is an autosomal-dominant hereditary cancer syndrome with 100% penetrance; it is characterized by the early onset of a large number of colorectal adenomas that invariably lead to colorectal carcinogenesis development before the age of 40 years (7). Thus, the development of effective chemopreventive interventions for these patients is important. In FAP clinical trials, celecoxib reduced the colorectal adenoma burden in approximately 50% of patients (8). NSAIDs were initially believed to act by inhibiting prostaglandin synthesis; however, various reports have questioned this concept, including one showing that PGE2 inhibition is not correlated with the reduction in polyp burden by celecoxib in patients with FAP (9). Thus, more predictive mechanistic biomarkers of NSAIDs' chemopreventive effects are still needed.

Lipoxygenases (LOX) are a class of enzymes that are involved in polyunsaturated fatty acid metabolism. Within this family,

[1]Departments of Palliative, Rehabilitation, and Integrative Medicine, The University of Texas MD Anderson Cancer Center, Houston, Texas. [2]Clinical Cancer Prevention, The University of Texas MD Anderson Cancer Center, Houston, Texas. [3]Gastrointestinal Medical Oncology, The University of Texas MD Anderson Cancer Center, Houston, Texas. [4]Department of Oncology, Lombardi Comprehensive Cancer Center, Georgetown University, Washington, D.C. [5]Department of Epidemiology, School of Public Health, The University of Texas Health Science Center, Houston, Texas.

Note: Supplementary data for this article are available at Cancer Prevention Research Online (http://cancerprevres.aacrjournals.org/).

Current address for I. Shureiqi: Division of Hematology and Oncology, Department of Internal Medicine, Rogel Cancer Center, University of Michigan, Ann Arbor, Michigan.

Corresponding Authors: Peiying Yang, Department of Palliative, Rehabilitation, and Integrative Medicine, The University of Texas MD Anderson Cancer Center, Houston, TX 77030. Phone: 713-563-7531; E-mail: pyang@mdanderson.org; and Imad Shureiqi, Division of Hematology and Oncology, Department of Internal Medicine, Rogel Cancer Center, University of Michigan, Ann Arbor, MI 48109. Phone: 734-647-1417; E-mail: ishureiq@med.umich.edu

Cancer Prev Res 2022;15:217–24

doi: 10.1158/1940-6207.CAPR-21-0066

15-lipoxygenase-1 (15-LOX-1) is the main enzyme that metabolizes linoleic acid, the predominant polyunsaturated fatty acid in the human diet, to produce 13-*S*-hydroxyoctadecadienoic acid (13-S-HODE), which has differential antitumorigenic effects compared with those of other LOX products (10). 15-LOX-1 is downregulated in human-invasive colorectal carcinogenesis and in colorectal adenomas in patients with FAP and non-FAP (11, 12). Celecoxib upregulates 15-LOX-1 expression and increases 13-S-HODE production to suppress colorectal cancer in preclinical models (13). However, whether celecoxib modulates the polyunsaturated fatty acid metabolism by LOXs to exert chemopreventive effects clinically remains unclear. In this study, we examined whether celecoxib modulates the polyunsaturated fatty acid metabolism via LOXs in the colon to affect its chemopreventive effects in patients with FAP.

Methods

Clinical study and human colonic tissue sampling

The Institutional Review Board (IRB) of The University of Texas MD Anderson Cancer Center (Houston, TX) approved our clinical protocol (NCT00503035), which ensured that studies are conducted in accordance with the recognized ethical principles (e.g., Declaration of Helsinki, Belmont Rule, U.S. Common Rule). For all patients, we obtained an institutional IRB-approved individual written informed consent from each subject before participating in the study. This single-arm celecoxib study enrolled 47 FAP patients at MD Anderson between November 2004 and May 2010. Twenty-seven patients completed the procedures required for the reported laboratory analyses. A baseline colonoscopy (or sigmoidoscopy in patients who had undergone colectomy) was performed before the initiation of celecoxib, and a follow-up colonoscopy or sigmoidoscopy was performed after celecoxib treatment. The celecoxib dose was 400 mg, given orally twice daily for 6 months. Patients' demographic and clinical characteristics, treatment schedules, and polyp burden assessment to determine response have been described previously (14). Celecoxib levels in sera were measured and assessed prior to the initiation of the study and then after 2, 4, and 6 months of treatment at the MD Anderson diagnostic laboratory or Quest Diagnostics.

Celecoxib treatment of $Apc^{\Delta 580}$ mice

Mouse care and experimental protocols were approved and conducted in accordance with the guidelines of the Animal Care and Use Committee of MD Anderson Cancer Center (Houston, TX). $Apc^{\Delta 580}$-flox mice were bred with CDX2-cre mice that specifically expressed Cre recombinase in the colon to generate $Apc^{\Delta 580}$-$flox^{(+/-)}$; $CDX2\text{-}Cre^{(+/-)}$ mice [$Apc^{\Delta 580(+/-)}$, designated as $Apc^{\Delta 580}$ mice in this study], in which one allele has an *Apc* codon 580 frame-shift mutation in colons.

Celecoxib powder was obtained from commercially available celecoxib capsules (200 mg/capsule, NDC 69097–421–07, Cipla USA, Inc.) and mixed with 4% carboxymethylcellulose sodium (catalog no. C9481, Sigma) to create a celecoxib suspension at a concentration of 20 mg/mL. We orally administered the celecoxib suspension via gavage to 14-week-old $Apc^{\Delta 580}$ mice with existing colon tumors ($n = 6$; ref. 15) at a final dose of 200 mg/kg/day for 5 consecutive days. The mice were euthanized, and the colonic tumors and adjacent normal colonic tissues were harvested, immediately flash-frozen in liquid nitrogen, and stored in a -80°C freezer until they were analyzed.

LC/MS/MS of celecoxib levels and LOXs' products

Lipid products and celecoxib were extracted in a manner similar to previously published methods (16). In brief, each frozen biopsy tissue sample was sectioned into 1- to 2-mm strips and were transferred to microcentrifuge tubes. An aliquot of 500 μL cold tissue homogenization buffer was then added to the samples before they were homogenized in an Ultrasonic Processor (Misonix) for 3.5 minutes (2x) at 0°C (17). Following centrifugation at 10,000 rpm for 5 minutes at 4°C, an aliquot of 400-μL supernatant was transferred to a glass tube and 600 μL of PBS buffer [1 mmol/L EDTA, 1% butylated hydroxytoluene and 10 μL of 12-, or 15-hydroxyeicosatetraenoic acid (12- or 15-HETE); leukotriene B_4 (LTB_4); or 13-HODE; 100 ng/mL] was added. Samples were acidified with 0.5 N HCl (pH = 3.2 to 3.3) and subjected to the extraction with 2 mL of ethyl acetate. The upper organic layer was collected and extraction was repeated two more times. The organic layers from three extractions were pooled and evaporated to dryness under a stream of nitrogen. Samples were then reconstituted in 100 μL of methanol and ammonium acetate buffer (10 mmol/L at pH 8.5; 70:30, v/v) before analyzed by LC/MS-MS). The concentration of protein was measured using a Bradford protein assay (Bio-Rad). Analysis of eicosanoids was performed using a Quattro Ultima tandem mass spectrometer (Waters) equipped with an Agilent HP 1100 binary-pump HPLC inlet, as described previously (16, 18). The lower limit of quantification was 0.39 ng/mL for LTB4, and 0.78 ng/mL for 12-HETE, 15-HETE, and 13-HODE. The lowest detection limit of these lipoxygenase products was 0.05–0.1 ng/mL when the signal to noise ratio was >3. The LC/MS-MS analysis for celecoxib was carried out under a similar condition as described above for the eicosanoid analysis, with some minor changes. In brief, 10 μL of the sample were injected to a Luna 3-μm phenyl-hexyl (2 × 150-mm) analytical column (Phenomenex). Celecoxib was detected and quantified by mass spectrometer using electrospray-negative ion mode and the transition m/z 380.2 > 316.1 for celecoxib was monitored. The results are shown as ng/mg protein.

An aliquot (100 μL) of serum was diluted with an equal volume of 10 mmol/L ammonium acetate, pH 8.5, prior to be extracted with 4 mL of hexane: ethyl acetate (1:1, v/v). The mixture was vortexed for 5 minutes and then centrifuged at 4,000 rpm for 5 minutes at 5°C. The extraction was repeated two more times, and the upper organic phase was collected, pooled, and evaporated to dryness under a stream of nitrogen. The sample was reconstituted in 200 μL of methanol: 10 mmol/L ammonium acetate, pH 8.5 (1:1, v/v). The level of

celecoxib in the samples was determined by LC/MS-MS. Serum celecoxib was quantified using authentic celecoxib standard. The results are presented as ng/mL.

To determine celecoxib levels in mouse colon tissues, we added an aliquot of 300 μL of PBS to each tissue sample and homogenized the tissues in screw capped microcentrifuge tubes with ceramic beads using the Precellys tissue homogenizer (Bertin Technologies). An aliquot of 200 μL of supernatant was mixed with 1 ml of methanol, vortexed, and centrifuged at 14,800 rpm at 4°C for 10 minutes. The supernatant was transferred and dried with nitrogen. Samples were then reconstituted with 100 μL of methanol and 0.1% formic acid in water (1:1) prior to LC/MS-MS analysis.

The analyses for celecoxib in mouse colon tissues were performed using a 6460 Triple Quad tandem mass spectrometer (Agilent Technologies) equipped with an Agilent HP 1200 binary-pump HPLC inlet. Celecoxib was analyzed on a Kinetex C_{18} column (2.6 μm, 2.0 × 100 mm; Phenomenex) using a gradient elution of 0.1% formic acids in water and 0.1% formic acid in acetonitrile. The column temperature was set at 35°C, and the flow rate was 0.4 mL/minute with a 10 minutes of total run time. The injection volume was 5 μL. Celecoxib was detected and quantified by the mass spectrometer operated in electrospray-negative ion mode and the transition *m/z* 380 > 316 was used to monitor the celecoxib, as described previously.

Diet, supplement and medication intake

A 137-item semi-quantitative food frequency questionnaire (FFQ) was used to assess dietary and alcohol intake. The FFQ prompted participants to report their intake over the previous 6 months (19). Participant data were entered into the Food Frequency Data Entry and Analysis Program to calculate the intake of 49 nutritional components as well as individual fatty

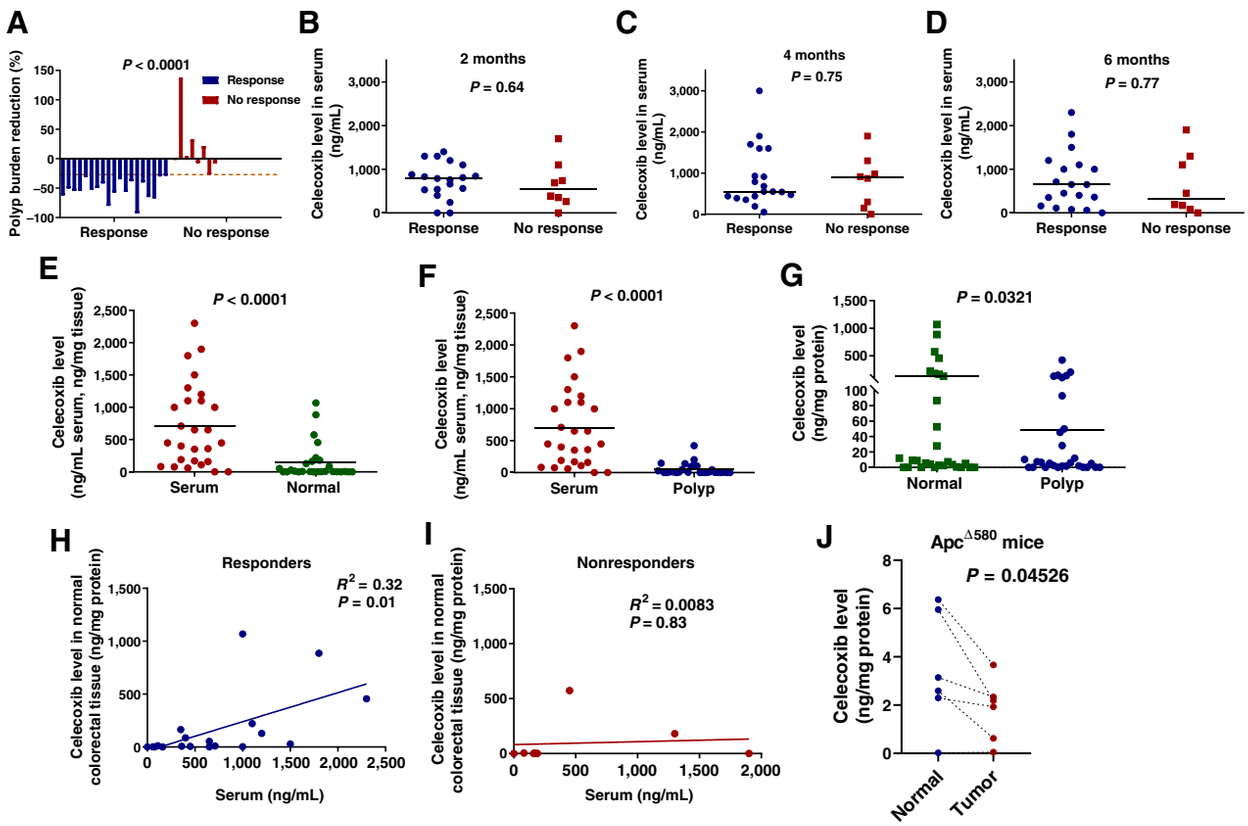

Figure 1.

Celecoxib levels in colorectal tissues and sera in humans and mice. **A–I,** Patients with FAP ($n = 27$) received 400 mg of oral celecoxib twice per day for 6 months. Colonoscopy was performed to assess polyp burden and collect colorectal tissue samples before and 6 months after celecoxib treatment. **A,** Celecoxib response statuses for each patient, based on the response cutoff of polyp tumor burden reduction by ≥ 28% from the baseline (brown dotted line). **B–D,** Comparisons of celecoxib levels in sera at 2 (**B**), 4 (**C**), and 6 (**D**) months of celecoxib treatment in patients who did and did not experience a celecoxib response. Lines represent medians. **E–G,** Comparisons of celecoxib levels in paired sera and normal colorectal tissues (**E**), paired sera and polyps (**F**), or paired normal colorectal tissues and polyps at six months of celecoxib treatment (**G**). Lines represent means. Correlation analyses of celecoxib levels between sera and normal colorectal tissues at 6 months of celecoxib treatment in the 19 of 27 FAP patients who experienced a response (**H**) and in the 8 of 27 patients with FAP who did not experience a response (**I**). **J,** Comparison of celecoxib levels in normal and polyp colorectal tissue samples from $Apc^{\Delta 580}$ mice. 14-week-old $Apc^{\Delta 580}$ mice received celecoxib (200 mg/kg/day) via gavage for 5 consecutive days ($n = 6$ mice) and then were euthanized. Celecoxib levels of the paired colorectal tumor and their adjacent normal colorectal tissues are shown.

acids. Nutrient and gram weight information for the FFQ items was obtained from the Food Intake Analysis System USDA Survey Nutrient Data Base [U.S. Department of Agriculture, Agricultural Research Service, 1997; ON: Nutrient Database for Individual Intake Surveys; 1994–1996 Continuing Survey of Food Intakes by Individuals, and 1994–1996 Diet and Health Knowledge Survey (CD-ROM)]. Intake of hormone replacement therapy, vitamins, and other nutritional supplements was measured using a medication questionnaire.

Statistical analysis

For paired two-group comparisons (e.g., normal tissues and polyps from the same subject), we used a paired *t* test. ANOVA (one-way) with Bonferroni adjustments was used for multiple group comparisons. All tests were two-sided, and significance was defined as $P < 0.05$. Data were analyzed using GraphPad Prism software 8.0.0 (GraphPad Software).

Results

Patients' demographic and clinical characteristics, treatment schedules and polyp burden assessments were described previously (14). Adherence to celecoxib intake was very high (14). Celecoxib responders were defined as those who experienced a ≥28% reduction in polyp tumor burden of the baseline (**Fig. 1A**; ref. 14). Nineteen of 27 patients were responders and 8 were nonresponders. All but two of the 27 subjects continued celecoxib treatment until their blood collections and colonoscopy procedures were performed at the end of treatment. One patient in the responders' group took the last dose

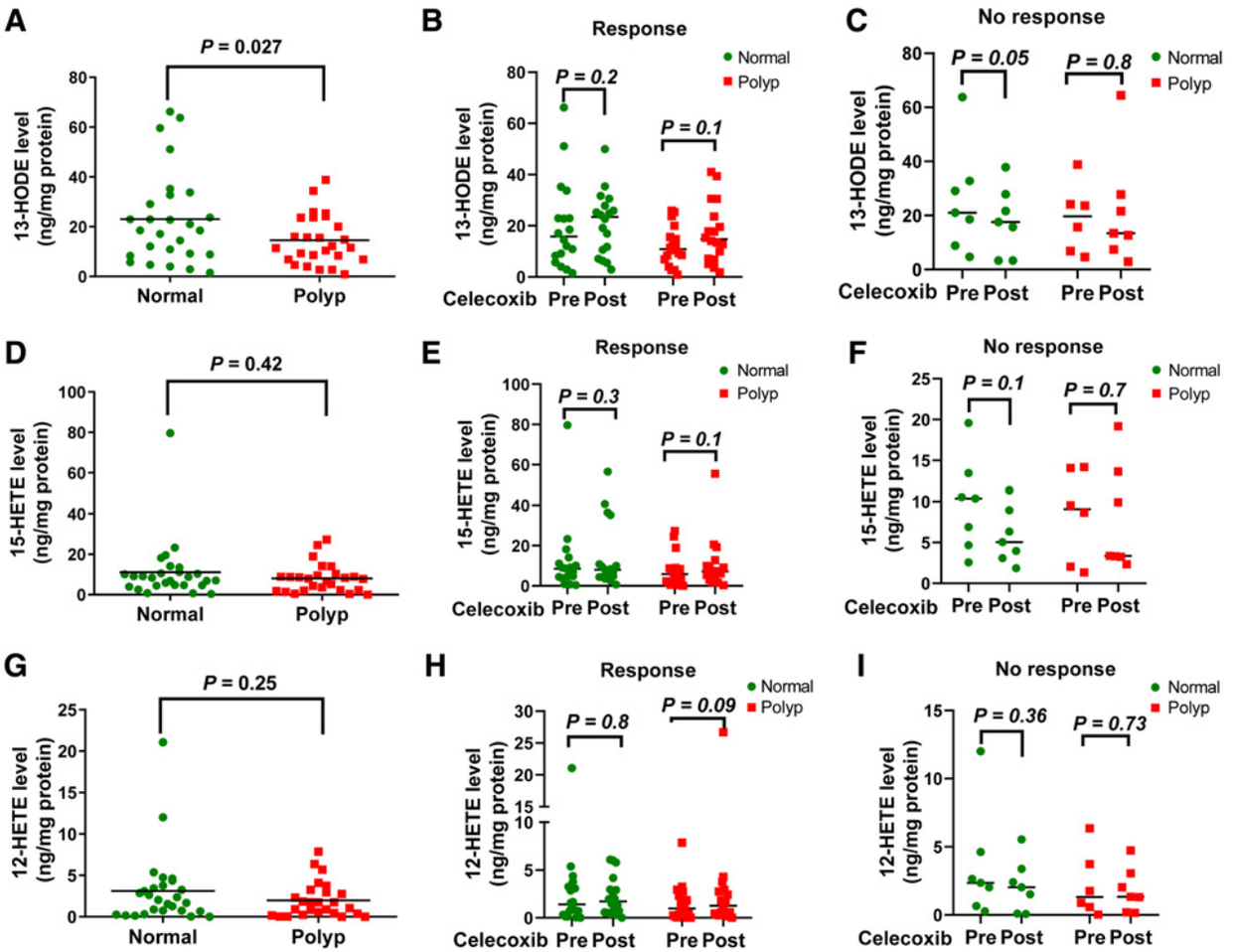

Figure 2.

Effects of celecoxib on generations of colorectal lipoxygenase products in patients with FAP. The indicated lipoxygenase products were measured by LC/MS-MS in normal tissues and polyps before and 6 months after celecoxib treatment. **A**, **D**, and **G,** Comparisons of 13-HODE (**A**), 15-HETE (**D**), and 12-HETE (**G**) levels in paired normal and polyp colorectal tissue samples from patients with FAP prior to celecoxib treatment ($n = 27$). Lines represent means. **B**, **C**, **E**, **F**, **H**, and **I,** Comparisons of 13-HODE (**B** and **C**), 15-HETE (**E** and **F**), and 12-HETE (**H** and **I**) levels between pre- and post- 6 months of celecoxib treatment in normal and polyp colorectal tissues of patients with FAP who did ($n = 18$) and did not ($n = 7$) experience a celecoxib response. Lines represent medians.

3 days prior to sample collection, and a patient in the non-responders' group took the last dose 6 days prior to sample collection, at the endpoints described above. Unlike the other 25 subjects, these two cases had undetectable celecoxib levels in their sera on the day of colonoscopy. Their oxylipin endpoint tissue results were not included in the oxylipin analyses because they might not be reflective of celecoxib's effects. Responders and nonresponders were similar in regard to age, sex, and BMI (Supplementary Table S1). Dietary intake between responders and non-responders did not significantly differ, including total fat and arachidonic and linoleic acids (Supplementary Table S1).

To assess whether celecoxib bioavailability affected clinical response, we measured celecoxib levels in sera and colorectal tissues. Celecoxib levels in sera were similar at 2, 4, and 6 months of celecoxib treatment between responders and nonresponders (**Fig. 1B–D**). Celecoxib levels were significantly lower in normal and polyp colorectal tissues than in their paired serum levels (**Fig. 1E** and **F**). Celecoxib levels (mean ± SEM) were significantly lower in polyps (48.99 ± 16.89 ng/mg protein) than in normal colorectal tissues (134.70 ± 50.51; **Fig. 1G**). Celecoxib levels in normal colorectal tissues and polyps tended to be higher in the responders' group than in the nonresponders' group, but the differences did not reach statistical significance (Supplementary Fig. S1A and S1B). The correlation of celecoxib levels in sera and normal colorectal tissues was stronger than in sera and polyps (Supplementary Fig. S1C and S1D). Furthermore, celecoxib levels in normal colorectal tissues were significantly correlated with those in sera in the responders (**Fig. 1H**), but not in the nonresponders (**Fig. 1I**). Celecoxib levels were also significantly lower in colorectal tumors (1.80 ± 0.52 ng/mg protein) than in paired normal colorectal tissues (3.40 ± 0.99) in celecoxib-treated $Apc^{\Delta 580}$ mice (**Fig. 1J**; Supplementary Fig. S2).

To evaluate celecoxib's effects on the oxidative metabolism of LOXs-mediated linoleic and arachidonic acids, we measured major LOX oxidative products 13-HODE, 15-HETE, 12-HETE, and LTB4 levels in colorectal tissues. 13-HODE levels were higher than 15-HETE, 12-HETE, and LTB4 levels in both normal colorectal tissues and polyps (Supplementary Fig. S3A and S3B) and higher in normal colorectal tissues than in polyps (mean ± SEM: 23.03 ± 3.57 vs. 14.56 ± 1.95 ng/mg protein; **Fig. 2A**), while 15-HETE and 12-HETE levels were similar between normal and polyps (**Fig. 2D** and **G**). LTB4 was lower than the detection limit in 44%-46% of the tissue samples. The detectable LTB4 levels were similar between normal and polyps (median = 0.07 ng/mg protein for normal tissue and 0.07 ng/mg protein for polyps; Supplementary Fig. S3C).

13-HODE levels after 6 months of celecoxib treatment tended to be higher than those from before treatment in both normal and polyp colorectal tissues in the responders' group, but these differences failed to reach statistical significance (**Fig. 2B**). In contrast, 13-HODE levels after celecoxib treatment tended to be lower than those before treatment in normal colorectal tissues and polyps from the nonresponders' group (**Fig. 2C**). The ratios of 13-HODE levels of 6 months of celecoxib treatment over pre-celecoxib treatment were significantly higher in normal colorectal tissues in responders than in those from non-responders ($P = 0.01$, Supplementary Fig. S4A); we also found a similar trend, although non-statistically significant, in the polyps of responders versus nonresponders ($P = 0.44$; Supplementary Fig. S4B). The levels of pre- and post-celecoxib treatments and the ratios of post- over pre-celecoxib treatment in normal colorectal tissues and polyps from responders and nonresponders were similar for 15-HETE (**Fig. 2E** and **F**; Supplementary Fig. S4C and D), 12-HETE (**Fig. 2H** and **I**; Supplementary Fig. S4E and S4F), and detectable LTB4 levels (Supplementary Fig. S4G and S4H).

Discussion

We found that celecoxib bioavailability was reduced in colorectal polyps. Both nonresponders and responders had good celecoxib systemic bioavailability, as measured by serum levels at 2, 4, and 6 months after celecoxib treatment. However, celecoxib levels were significantly lower in polyps than in paired normal colorectal tissues in patients with FAP. The lack of correlation in celecoxib levels between sera and normal colorectal tissues in nonresponders further suggests that reduced bioavailability in colorectal tissues contributed to the failure of celecoxib to reduce polyp formation. Studies in $Apc^{\Delta 580}$ mice that experimentally model human FAP similarly showed that celecoxib levels were lower in colorectal tumors than in their paired normal-appearing colorectal tissues. This observation further supports the concept that the reduction of celecoxib bioavailability limits the chemopreventive effectiveness of celecoxib.

Celecoxib undergoes extensive hepatic metabolism in humans primarily via the cytochrome P450 2C9 (CYP2C9) enzyme to form hydroxycelecoxib, which is further oxidized to form carboxycelecoxib and then conjugated with glucuronic acid by UGT to form the 1-O-glucuronide that is excreted in feces and urine (20). *CYP2C9* polymorphism significantly modulates celecoxib's pharmacokinetics and pharmacodynamics (21). For example, 400 mg of celecoxib reduces adenoma recurrence risk more than does the 200 mg dose only in individuals with the *CYP2C9*3* genotype variant, while celecoxib's cardiovascular side effects are increased at 200 mg dose in subjects with wild-type but not in those with *CYP2C9*2* or *CYP2C9*3* variants (22). In our small study, we did not measure *CYP2C9* genotypes because of the low frequency of these genotype variants (6%–10% in general population) (22). Nevertheless, serum celecoxib levels at the time of response assessment were similar between responders and nonresponders, which strongly suggests that our response differences were unlikely to have been affected by *CYP2C9* genotype variants. The differential bioavailability of celecoxib in the normal colon and colorectal polyps could be mediated by somatic CYP2C9 expression differences between normal and polyp tissues, as

CYP2C9 expression levels have been reported to be higher in colon adenocarcinoma than in paired normal colonic mucosa (23). Alternatively, the differentially reduced bioavailability of celecoxib in colorectal polyps may be the result of differential vascular delivery between normal colons and polyps. In advanced cancer, reduced drug bioavailability in cancer tissues has been well described and documented to limit anticancer treatment activity (24, 25). Whether this principle applies to earlier stages of tumorigenesis is poorly understood. Further investigation is thus warranted to determine whether CYP2C9 is differentially expressed in polyps or whether dysfunctional vasculature in the polyps modulates celecoxib bioavailability. Meanwhile, our data demonstrate celecoxib's differential bioavailability in colorectal polyps, suggesting that monitoring levels of antitumorigenic agents in tissues rather than in sera is needed for better chemopreventive response prediction.

Our results confirmed that the reduction in 13-HODE in colorectal polyps differed compared with that in other LOX products, and was unrelated to the difference in the intake of its substrate (i.e., linoleic acid) or other dietary factors (16). 13-HODE levels in normal colorectal tissues, and to a lesser degree in polyps, tended to increase in celecoxib responders after 6 months of celecoxib treatment but tended to decrease in nonresponders, although these differences were statistically non-significant. Nonetheless, the changes in normal colorectal tissues' 13-HODE levels (ratios of post- over pre-celecoxib treatment) were significantly different between responders and nonresponders. Our inability to detect a statistically difference in celecoxib modulation of 13-HODE in patients with FAP who experienced a response to celecoxib could be related to (i) the small sample size; and (ii) low levels of celecoxib in colorectal tissues at the time of measurement [9/19 subjects in the responder group had celecoxib levels less than 50 ng/mg (approximately 0.13 μmol/L) protein in normal colorectal tissues]. These celecoxib levels are significantly lower than the lowest reported celecoxib level (12.5 μmol/L) to induce 15-LOX-1 expression and increase 13-HODE levels in preclinical studies (13). Our findings nevertheless underscore the need to measure antitumorigenic drugs' concentrations in target tissues when assessing the effects of these agents on putative biomarkers to ensure adequate drug concentrations for targeting the biomarker of interest.

One of our study limitations is that our population was restricted to patients with FAP; there might be differences in celecoxib mechanisms between patients with sporadic colorectal carcinogenesis and FAP. Further studies, including in patients with sporadic colorectal carcinogenesis, are needed to assess the potential mechanisms of this reduced celecoxib bioavailability to identify approaches to enhance chemopreventive activity.

In conclusion, our findings demonstrate the need to consider the bioavailability of chemo-preventive agents in target tissues as a potential rate-limiting factor to their effectiveness and the need to identify strategies to address this resistance mechanism.

Authors' Disclosures

P. Yang reports grants from NCI during the conduct of the study. I. Shureiqi reports grants from NIH/NCI during the conduct of the study; grants from NIH/NCI, and grants from Cancer Prevention and Research Institute of Texas outside the submitted work. X. Zuo reports grants from NIH/NCI during the conduct of the study. No disclosures were reported by the other authors.

Authors' Contributions

P. Yang: Formal analysis, funding acquisition, methodology, writing–review and editing. **X. Zuo:** Formal analysis, investigation, methodology, writing–review and editing. **S. Advani:** Formal analysis, writing–original draft. **B. Wei:** Investigation, methodology. **J. Malek:** Investigation. **R.S. Day:** Formal analysis, writing–review and editing. **I. Shureiqi:** Conceptualization, data curation, formal analysis, supervision, funding acquisition, investigation, writing–original draft, project administration, writing–review and editing.

Acknowledgments

This work was supported by the National Cancer Institute (R01-CA106577 and R01-CA137213, to I. Shureiqi, R01-CA144053, to P. Yang, and R03-CA235106 to X. Zuo). We acknowledge all of our patients, who contributed their time and valuable resources to this research.

Received February 11, 2021; revised June 14, 2021; accepted September 23, 2021; published first October 5, 2021.

References

1. Thun MJ, Henley SJ, Patrono C. Nonsteroidal anti-inflammatory drugs as anticancer agents: mechanistic, pharmacologic, and clinical issues. J Natl Cancer Inst 2002;94:252–66.
2. Janne PA, Mayer RJ. Chemoprevention of colorectal cancer. N Engl J Med 2000;342:1960–8.
3. Sandler RS, Halabi S, Baron JA, Budinger S, Paskett E, Keresztes R, et al. A randomized trial of aspirin to prevent colorectal adenomas in patients with previous colorectal cancer. N Engl J Med 2003;348:883–90.
4. Baron JA, Cole BF, Sandler RS, Haile RW, Ahnen D, Bresalier R, et al. A randomized trial of aspirin to prevent colorectal adenomas. N Engl J Med 2003;348:891–9.
5. Solomon DH, Schneeweiss S, Glynn RJ, Kiyota Y, Levin R, Mogun H, et al. Relationship between selective cyclooxygenase-2 inhibitors and acute myocardial infarction in older adults. Circulation 2004;109:2068–73.
6. Mamdani M, Rochon P, Juurlink DN, Anderson GM, Kopp A, Naglie G, et al. Effect of selective cyclooxygenase 2 inhibitors and naproxen on short-term risk of acute myocardial infarction in the elderly. Arch Intern Med 2003;163:481–6.
7. Dunlop M. Guidance on gastrointestinal surveillance for hereditary non-polyposis colorectal cancer, familial adenomatous polypolis, juvenile polyposis, and Peutz-Jeghers syndrome. Gut 2002;51:v21–v7.
8. Steinbach G, Lynch PM, Phillips RK, Wallace MH, Hawk E, Gordon GB, et al. The effect of celecoxib, a cyclooxygenase-2 inhibitor, in familial adenomatous polyposis. N Engl J Med 2000;342:1946–52.
9. Sinicrope FA, Half E, Morris JS, Lynch PM, Morrow JD, Levin B, et al. Cell proliferation and apoptotic indices predict adenoma regression in

a placebo-controlled trial of celecoxib in familial adenomatous polyposis patients. Cancer Epidemiol Biomarkers Prev 2004;13:920–7.
10. Shureiqi I, Lippman SM. Lipoxygenase modulation to reverse carcinogenesis. Cancer Res 2001;61:6307–12.
11. Shureiqi I, Wojno KJ, Poore JA, Reddy RG, Moussalli MJ, Spindler SA, et al. Decreased 13-S-hydroxyoctadecadienoic acid levels and 15-lipoxygenase-1 expression in human colon cancers. Carcinogenesis 1999;20:1985–95.
12. Zuo X, Shureiqi I. Eicosanoid profiling in colon cancer: Emergence of a pattern. Prostaglandins Other Lipid Mediat 2013;104–105:139–43.
13. Shureiqi I, Jiang W, Zuo X, Wu Y, Stimmel JB, Leesnitzer LM, et al. The 15-lipoxygenase-1 product 13-S-hydroxyoctadecadienoic acid down-regulates PPAR-delta to induce apoptosis in colorectal cancer cells. Proc Natl Acad Sci U S A 2003;100:9968–73.
14. Lynch PM, Morris JS, Ross WA, Rodriguez-Bigas MA, Posadas J, Khalaf R, et al. Global quantitative assessment of the colorectal polyp burden in familial adenomatous polyposis by using a Web-based tool. Gastrointest Endosc 2013;77:455–63.
15. Liu Y, Deguchi Y, Tian R, Wei D, Wu L, Chen W, et al. Pleiotropic Effects of PPARD Accelerate Colorectal Tumorigenesis, Progression, and Invasion. Cancer Res 2019;79:954–69.
16. Shureiqi I, Chen D, Day RS, Zuo X, Hochman FL, Ross WA, et al. Profiling lipoxygenase metabolism in specific steps of colorectal tumorigenesis. Cancer Prev Res 2010;3:829–38.
17. Vanamala J, Glagolenko A, Yang P, Carroll RJ, Murphy ME, Newman RA, et al. Dietary fish oil and pectin enhance colonocyte apoptosis in part through suppression of PPARdelta/PGE2 and elevation of PGE3. Carcinogenesis 2008;29:790–6.
18. Yang P, Chan D, Felix E, Madden T, Klein RD, Shureiqi I, et al. Determination of endogenous tissue inflammation profiles by LC/MS/MS: COX- and LOX-derived bioactive lipids. Prostaglandins Leukot Essent Fatty Acids 2006;75:385–95.
19. Martinez ME, McPherson RS, Annegers JF, Levin B. Association of diet and colorectal adenomatous polyps: dietary fiber, calcium, and total fat. Epidemiology 1996;7:264–8.
20. Davies NM, McLachlan AJ, Day RO, Williams KM. Clinical pharmacokinetics and pharmacodynamics of celecoxib: a selective cyclooxygenase-2 inhibitor. Clin Pharmacokinet 2000;38:225–42.
21. McEvoy L, Carr DF, Pirmohamed M. Pharmacogenomics of NSAID-Induced Upper Gastrointestinal Toxicity. Front Pharmacol 2021;12:684162.
22. Chan AT, Zauber AG, Hsu M, Breazna A, Hunter DJ, Rosenstein RB, et al. Cytochrome P450 2C9 variants influence response to celecoxib for prevention of colorectal adenoma. Gastroenterology 2009;136:2127–36.
23. Enayetallah AE, French RA, Grant DF. Distribution of soluble epoxide hydrolase, cytochrome P450 2C8, 2C9 and 2J2 in human malignant neoplasms. J Mol Histol 2006;37:133–41.
24. Tannock IF, Lee CM, Tunggal JK, Cowan DSM, Egorin MJ. Limited penetration of anticancer drugs through tumor tissue. Clin Cancer Res 2002;8:878.
25. Minchinton AI, Tannock IF. Drug penetration in solid tumours. Nat Rev Cancer 2006;6:583–92.