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NCI, DCPC
Chemoprevention Branch and Agent Development Committee

# CLINICAL DEVELOPMENT PLAN: SULINDAC SULFONE

## DRUG IDENTIFICATION

**CAS Registry No.:** 59864-04-9

**CAS Name (9CI):** (Z)-5-Fluoro-2-methyl-1-((4-methylsulfonyl)phenylmethylene)-1*H*-indene-3-acetic Acid

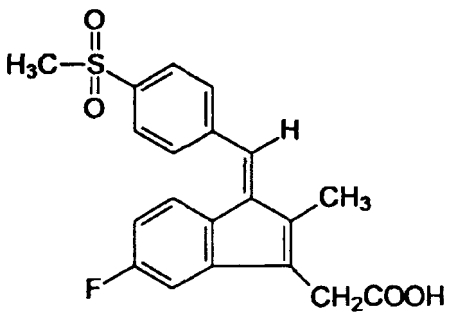
**Synonyms:** FGN-1

**Related Compounds:**

Sulindac
(Z)-5-Fluoro-2-methyl-1-((4-methylsulfinyl)phenylmethylene)-1*H*-indene-3-acetic Acid
Clinoril® (Active Ingredient)
Sulindac Sulfide
(Z)-5-Fluoro-2-methyl-1-(*p*-methylthiobenzylidene)-3-indenylacetic Acid

**Molecular Wt.:** 372

**Structure:**




## EXECUTIVE SUMMARY

The NCI, Chemoprevention Branch is developing sulindac sulfone as a chemopreventive agent under a Clinical Trial Agreement with Cell Pathways, Inc. (CPI; Denver, CO). Sulindac sulfone is a metabolite of the FDA-approved nonsteroidal antiinflammatory drug (NSAID) sulindac, also currently under development by the Chemoprevention Branch [1]. Sulindac, the parent compound, has been shown to be effective in preventing carcinogen-induced rat colon and mouse bladder cancer [2,3] and in causing the regression of colorectal adenomas in cases of familial adenomatous polyposis (FAP) [4–6] or Gardner's syndrome [7], a phenotypic variant of FAP. FAP is a hereditary disease characterized by the occurrence of hundreds to thousands of adenomatous polyps in the colon. Adenomatous polyps are considered to be intermediates in the pathologic progression from normal colorectal epithelia to carcinomas in both sporadic and familial settings. Colorectal adenomas, or subsets of them defined on the basis of their size or histopathology, are considered validated and predic-

tive intermediate biomarkers of cancer risk worthy of surgical interventions such as polypectomy. The potential of medical interventions to regress preinvasive neoplasia or to moderate the cancerous progression of preinvasive neoplasia forms the basis for clinical chemoprevention trials in subjects demonstrating this histopathology at baseline.

Like the parent compound, sulindac sulfone has shown activity in preventing carcinogen-induced tumors in rats. In preclinical efficacy studies, dietary sulindac sulfone inhibited colonic carcinogenesis in AOM-treated rats. Sulindac sulfone treatment caused a reduction in both tumor burden and multiplicity and increased tumor latency, although a higher concentration of sulindac sulfone was required to achieve tumor inhibitory activity similar to that of sulindac. In another study, dietary sulindac sulfone treatment was shown to be effective against MNU-induced mammary tumors in rats. Sulindac sulfone also inhibited growth of colon cancer cell lines and excised human colonic polyps *in vitro*. This evidence is sufficient to warrant clinical development of sulindac sulfone as a cancer chemopreventive drug.

As stated above, sulindac has prevented or caused the regression of colorectal adenomas in patients with FAP or Gardner's syndrome; however, polyp counts generally returned to pretreatment levels shortly after discontinuing treatment. Similarly, human epidemiologic studies have shown the duration of NSAID administration to be important in deriving protection from incident colorectal adenomas and cancer. Generally, use for less than 10 years is associated with no demonstrable efficacy. For these reasons, it is assumed that chronic administration of the drug will be necessary for effective prevention against colorectal neoplasia. Chronic exposure to NSAIDs is known to be associated with a variety of untoward effects including gastrointestinal ulceration which is believed to be a consequence of the NSAIDs inhibitory effects on prostaglandin synthesis; moreover, sulindac is believed to be one of the more toxic NSAIDs. In contrast, sulindac sulfone lacks significant antiprostaglandin synthesis activity at effective doses in animal models and therefore may offer a much better therapeutic index than classic NSAIDs for cancer chemoprevention.

One aspect of the development of cancer chemopreventive drugs by the NCI, Chemoprevention Branch is to identify and validate intermediate biomarkers as potential surrogate endpoints for cancer. As mentioned above, adenomatous polyps are premalignant histopathologic lesions used as histological intermediate biomarkers of colorectal carcinogenesis. Colorectal epithelial homeostasis is dependent on a balance between cell proliferation and programmed cell death (apoptosis) [8]. In FAP patients, the crypt proliferative zone shifts upwards, and apoptosis in the upper crypt decreases, resulting in the abnormal growth of this tissue [9]. NSAID interventions have been associated with variable effects on cellular proliferation [10–12], though in the setting of FAP, treatment with sulindac has been shown to induce apoptosis in the rectal mucosa [13]. Moreover, both the active metabolite of sulindac and sulindac sulfone were reported to inhibit the growth of colon cancer cell lines *in vitro* by inducing apoptosis [14–16]. Therefore, another potential intermediate biomarker of tumorigenesis would be the apoptotic index or perhaps ideally, an index of cellular population dynamics which integrates both proliferation and apoptosis into a single measure.

Preclinical toxicity studies of sulindac sulfone in mice, rats and dogs have been performed by CPI. In acute studies, rats exhibited only minimal gastrointestinal toxicity when dosed orally with high concentrations (1.8–6.0 mmol/kg-bw) of sulindac sulfone; however, lower doses were shown to increase the severity of stress-induced gastric ulcers in male rats.

An NCI, Chemoprevention Branch-sponsored Phase I clinical safety and pharmacokinetic study with sulindac sulfone is in progress. FAP patients with subtotal colectomies are being administered sulindac sulfone at 200, 300 and 400 mg bid (15.3, 23 and 30.6 μmol/kg-bw/day) for six months. Preliminary estimates of the efficacy of sulindac sulfone will be available in this study from serial evaluations of the number and size of colorectal adenomas. This study is also evaluating mechanisms of action of sulindac sulfone and intermediate biomarkers in FAP patients. CPI has conducted two Phase I safety and pharmacokinetic clinical studies on healthy male subjects under both fed and fasted conditions. The NCI, Chemoprevention Branch and CPI are considering Phase II clinical efficacy studies on patients with low-grade FAP and patients with sporadic colorectal adenomas. It is anticipated that the efficacy of sulindac sulfone on both breast and cervical neoplasia may be explored in future Phase II clinical trials.

CPI has filed a patent for sulindac sulfone and has agreed to supply the drug for the clinical studies.

## PRECLINICAL EFFICACY STUDIES

CPI has funded several studies investigating the effects of sulindac sulfone on chemically induced tumors in rats. One study reported the effects of sulindac sulfone on AOM-induced rat colon tumors. Sulindac sulfone resulted in a dose-dependent reduction in both tumor burden and tumor number in AOM-treated male Fisher-344 rats. Following AOM treatment, animals were administered sulindac sulfone in the daily diet for 31 weeks. Animals treated with 0.1% or 0.2% sulindac sulfone in the diet (134 or 269 μmol/kg-bw/day) resulted in 55% and 79% reductions, respectively, in tumor multiplicity when compared with AOM-treated control animals, although no comments were made regarding any toxicities seen with either of these doses [17]. This study determined that a five-fold higher concentration of sulindac sulfone was required to achieve the same inhibitory effect as seen with sulindac and a 13-fold higher concentration to achieve inhibitory activity similar to the NSAID piroxicam. A future opportunity for Chemoprevention Branch testing in another relevant rodent model is the transgenic Min mouse which harbors APC mutations and a phenotype reminiscent of FAP patients with multiple intestinal adenomas and accelerated neoplastic progression to invasive carcinoma [18]. NSAIDs have suppressed tumorigenesis in this model.

Sulindac sulfone was also reported to be effective in inhibiting mammary tumors in MNU-treated female Sprague-Dawley rats. Animals were treated with dietary 0.03% and 0.06% sulindac sulfone (*ca.* 40 and 80 μmol/kg-bw/day) starting seven days post-MNU. Sulindac sulfone significantly inhibited total tumor burden, incidence and multiplicity and was effective in increasing tumor latency [19]. The higher dose of sulindac sulfone was as effective as sulindac in reducing tumor size and multiplicity. In both of these studies, sulindac sulfone did not significantly inhibit prostaglandin synthesis at effective doses.

These results are supported by *in vitro* studies on human colon cancer cell lines. At the highest dose, sulindac sulfone inhibited the growth of the human colon cancer cell lines HT-29, DLD-1 and SW480 with reported $IC_{50}$ values of 119, 49 and 130 μM, respectively [20]. Approximately twice as much sulindac sulfone was required to achieve the same growth inhibitory activity as the active metabolite of the parent compound (sulindac sulfide). Sulindac sulfone has also been shown to inhibit the growth of epithelial cells from excised human colonic polyps. The growth inhibitory activity of sulindac sulfone on both the HT-29 cell line and the human breast cancer cell line MCF-7 was determined to be a result of increased programmed cell death (apoptosis) rather than a decrease in cellular proliferation [16,21].

## PRECLINICAL SAFETY STUDIES

*Safety*: There are no preclinical toxicity studies on sulindac sulfone completed or planned by the NCI, Chemoprevention Branch. CPI has performed a six-month toxicity study on rats and Beagle dogs and the NOEL has been determined. CPI has also determined the $LD_{50}$ for rats and mice when sulindac sulfone was given by either ig or ip administration. In acute studies in rats, only minimal gastrointestinal toxicity was detected when 670– 2,250 mg sulindac sulfone/kg-bw (1.8–6.0 mmol/kg-bw) was administered orally, although the severity of acute stress-induced gastric ulcers in male rats was increased in the presence of 4 mg sulindac sulfone/kg-bw (11 μmol/kg-bw) [22]. CPI has conducted full Segment I reproductive studies in rats and Segment II teratology studies in both rats and New Zealand White rabbits. Although no carcinogenicity studies have been performed for sulindac sulfone, Ames mutagenicity studies were performed on *Salmonella typhimurium* strains TA98, TA100, TA1537 and TA1538 and *Escherichia coli* strain WP2 uvrA.

*ADME*: There are no NCI, Chemoprevention Branch-funded preclinical pharmacokinetic studies planned for sulindac sulfone. CPI has conducted pharmacokinetic studies on both rats and Beagle dogs. They have identified gender differences in peak plasma levels ($C_{max}$) and plasma half life ($t_{½}$) for sulindac sulfone when administered orally and have determined the major routes of excretion.

## CLINICAL SAFETY: PHASE I STUDIES

An NCI, Chemoprevention Branch-sponsored six month Phase I clinical safety and pharmacokinetic study of sulindac sulfone is in progress (Dr. G. Thomas Budd, The Cleveland Clinic Foundation). This study is being performed on patients with FAP who have undergone a subtotal colectomy. This patient population was selected because of its clinical

relevance and sulindac's known efficacy in reducing adenoma size and number in FAP patients. In addition, it is essential to define ADME of sulindac sulfone in subjects with subtotal colectomies since it is known that the parent compound is reabsorbed from the gastrointestinal tract [1,23]. Two Phase I safety and pharmacokinetic studies on healthy male subjects have also been conducted by CPI.

*Drug Effect Measurement*: Since the mechanism of action of sulindac sulfone is not well defined, no definitive drug effect measurements have been identified. A decrease in the number and size of colorectal adenomas is being evaluated as a possible clinical marker of drug effect. In addition, studies are being performed to determine the effect of sulindac sulfone on potential cellular markers of drug effect including mucosal proliferation and on the apoptotic index of normal appearing mucosa and resected colorectal adenomas.

*Safety*: A six-month Chemoprevention Branch-sponsored Phase I clinical trial of sulindac sulfone is in progress. This study is designed to assess the safety of multiple oral doses of sulindac sulfone in FAP patients with subtotal colectomy and ileorectal anastamosis. Other objectives of this study are to obtain dose-ranging data, to determine the pharmacokinetics following a single oral dose and at steady state during twice daily dosing, and to investigate mechanisms of action. Patients were administered one of three doses (200, 300 or 400 mg bid) of sulindac sulfone orally for six months. Fifteen patients have entered the study and all six patients at the 200 mg bid dose (15.3 μmol/kg-bw/day) have completed the six-month study. CPI has arranged to extend the study for an undetermined period of agent administration.

In addition, two CPI-sponsored Phase I single dose safety and pharmacokinetic studies on sulindac sulfone have been completed in healthy male subjects.

*ADME*: CPI has completed two Phase I pharmacokinetic studies with sulindac sulfone. In one of these studies, which was recently published [24], healthy male subjects were given a single oral dose of sulindac sulfone ranging from 50 mg to 400 mg (2 to 16 μmol/kg-bw). Sulindac sulfone was detected in the plasma after a short lag time ($t_{lag}$) of approximately 0.25 hours. Peak plasma concentrations ($C_{max}$) were observed to roughly increase with increasing dose and ranged from 2 μg/ml to 13 μg/ml (5.4 to 35.1 μM), corresponding to the 50 mg and 400 mg dose, respectively. Peak plasma concentrations were reached by 1.5 to 3.0 hours ($t_{max}$) and the agent was essentially eliminated by 12 hours. Mean residence time in the plasma averaged 9 to 11 hours. The average urinary recovery was 30% of the oral dose. The fecal recovery varied greatly within dose groups, but averaged less than 10% of the oral dose and showed no apparent relationship with dosage [24].

## CLINICAL EFFICACY: PHASE II/III STUDIES

The NCI, Chemoprevention Branch and CPI are considering a Phase II clinical efficacy study in patients with low-grade FAP who have retained colorectal segments and have had previously resected colon adenomas with retained colorectal segments. This study will determine drug efficacy and identify potential intermediate biomarkers of colorectal tumorigenesis. Another clinical trial on subjects with sporadic adenomatous polyps is also under consideration. The NCI, Chemoprevention Branch and CPI are considering additional Phase II trials to investigate the efficacy of sulindac sulfone against breast and cervical neoplasia.

## PHARMACODYNAMICS

Sulindac sulfone was found to be effective in preventing colon tumors in AOM-treated rats at a dose of 270 μmol/kg-bw/day although it was not associated with significant reductions in prostaglandin levels in colorectal tissue. This dose was five-fold higher than that required to achieve the same level of inhibition as sulindac, the parent compound [17,25,26]. Clinical studies have shown that 150 mg bid (12 μmol/kg-bw/day) of sulindac is effective in inhibiting colorectal carcinogenesis in humans [4,6, 7,13]. Results from the Phase I multidose trial of sulindac sulfone will indicate whether a five-fold higher dose will be below the maximum safe dose. If this dose proves to be toxic, it may not be possible to achieve a safe yet efficacious dose when taken orally, though it is not known if the pharmacokinetics of sulindac sulfone are the same in rats and humans. Administration of sulindac sulfone by rectal suppositories might provide an efficacious dose while avoiding toxicity, a method of administration previously shown to be effective with the parent compound in FAP patients with colectomies [27].

## PROPOSED STRATEGY FOR CLINICAL DEVELOPMENT

### Drug Effect Measurement Issues

Drug effect measurements for sulindac sulfone have not been identified since the mode of action is not known. A decrease in the number and size of colorectal adenomas is being evaluated as a possible clinical drug effect marker. *In vitro* studies have shown that sulindac sulfone inhibits the growth of colon cancer cells by inducing apoptosis. Studies are in progress to evaluate the effect of sulindac sulfone on the apoptotic index of normal appearing mucosa and resected colorectal adenomas.

### Safety Issues

One problem with using sulindac as a cancer chemopreventive drug is the gastrointestinal toxicity resulting from chronic administration. This is believed to be a side effect from inhibition of prostaglandin synthesis. Unlike sulindac, sulindac sulfone lacks significant cyclooxygenase inhibitory activity, and thus may be much less toxic [19]. Studies should be performed to investigate the effects of chronic administration of sulindac sulfone on the upper gastrointestinal tract and other organs; preliminary estimations of upper gastrointestinal tract toxicities should be available from the Chemoprevention Branch-sponsored Phase I study.

### Pharmacodynamics Issues

Following a single oral dose of sulindac, humans excrete the majority of the sulfone metabolite through the urine; less than 10% of the oral dose is found in the feces [1]. It is unknown if sulindac sulfone taken orally would have a similar pattern of excretion, especially in FAP patients with subtotal colectomies, though this is being addressed in the Phase Ib clinical trial. It is also not known if sulindac sulfone exhibits a similar pattern of biliary excretion and enterohepatic circulation as seen with the parent compound [1] or if the pharmacokinetics are similar in patients with an intact colon. Depending on the outcome of these studies; it may be necessary to explore alternative means of drug delivery, such as time release encapsulation or rectal suppository.

### Regulatory Issues

Humans have been indirectly exposed to sulindac sulfone as a result of ingestion and metabolism of sulindac, although not at serum concentrations anticipated from the pharmacologic administration of sulindac sulfone itself, as in the NCI, Chemoprevention Branch Phase I study. Sulindac is an FDA-approved drug and the manufacturer (Merck, Sharp & Dohme) has performed extensive preclinical and clinical safety studies on this compound. CPI has performed a six-month toxicity study of sulindac sulfone in both rats and dogs. CPI has extended the six-month NCI, Chemoprevention Branch-sponsored Phase I clinical safety study on sulindac sulfone; depending on the length of the extension, it may be necessary to conduct additional preclinical safety studies.

### Intermediate Biomarker Issues

One goal of the development of cancer chemopreventive drugs by the NCI, Chemoprevention Branch is to identify and validate intermediate biomarkers as potential surrogate endpoints for cancer incidence. Adenomatous polyps are premalignant lesions and can be used as histopathological intermediate biomarkers of colorectal carcinogenesis. Colorectal epithelial homeostasis is dependent on a balance between cell proliferation and programmed cell death (apoptosis) [8]. In patients with colorectal adenomas, the crypt proliferative zone shifts upwards and apoptosis in this region decreases resulting in the abnormal growth of this tissue [9]. When these patients are treated with the NSAID sulindac, apoptosis in the rectal mucosa increases [13]. Moreover, sulindac sulfone was reported to inhibit the growth of colon cancer cell lines *in vitro* by inducing apoptosis [16]. Therefore, another important intermediate biomarker to be investigated on the cellular level is the determination of the apoptotic index [28].

In addition to histologic or cellular biomarkers, there are a number of molecular and/or genetic alterations associated with colon carcinogenesis [29,30]. These include alterations in DNA methylation patterns [31] and abnormalities in or altered expression of both tumor suppressor genes and oncogenes, including *bcl-2* [9,32–34], p53 [29,33, 35], *ras* [36,37] and *dcc* [38]. Determination of mutations, alterations in methylation patterns or other determinants of gene expression should therefore be explored as intermediate biomarkers for colorectal tumorigenesis.

FAP is an autosomal dominant inherited disease; afflicted individuals inherit a mutant allele of the *APC* gene [39]. It has been proposed that somatic mutations of the *APC* gene may be responsible for the

tions of the *APC* gene may be responsible for the phenotypic expression of tumorigenesis in FAP patients and be involved early in the development of sporadic adenomatous polyps as well [39,40]. Detection of *APC* mutations in patients with sporadic adenomatous polyps could be an early intermediate molecular biomarker for colorectal carcinogenesis.

Possible intermediate biomarkers for the proposed Phase II breast neoplasia efficacy trial include modulation of DCIS and other histological lesions, DNA ploidy, and proliferation markers (PCNA and Ki-67). For the proposed Phase II efficacy trial for cervical neoplasia, possible intermediate biomarkers include modulation of CIN lesions, DNA ploidy, micronucleated cell frequency, DNA content, PCNA, *ras* oncogene expression, EGFR, keratins and involucrin. In each instance, modulation of preinvasive neoplasia assumes the greatest priority among biomarkers under investigation in chemoprevention since it remains the most clinically relevant and validated measure of interventive efficacy. Indeed, histopathologic preinvasive neoplasia is already a validated marker of cancer risk in a variety of epithelial sites, necessitating surgical and/or radiologic interventions (*e.g.*, CIN, DCIS, colorectal adenomas, and high-grade esophageal dysplasia) [41–44].

### Supply and Formulation Issues

CPI holds a patent for sulindac sulfone and has entered into a Clinical Trial Agreement with the NCI, Chemoprevention Branch to pursue the development of this drug as a cancer chemopreventive agent. CPI has agreed to supply sulindac sulfone to the NCI, Chemoprevention Branch, therefore no supply problems are anticipated.

### Clinical Studies Issues

Sulindac has been shown to be very effective in reducing or preventing colorectal adenomas in patients with FAP. The desire to find a NSAID or NSAID metabolite with an improved therapeutic index led to the development of sulindac sulfone as a colorectal cancer chemopreventive drug. An NCI, Chemoprevention Branch-sponsored Phase Ib study of sulindac sulfone on FAP patients is nearing completion. This study was also designed to investigate the mechanisms of action of sulindac sulfone in FAP patients, to obtain preliminary indications of efficacy, and identify the maximum safe dose for subsequent Phase II investigations. In the future it may be necessary to explore the mechanism of action in a non-genetically predisposed patient population; indeed a Phase II clinical trial on subjects with sporadic colon polyps is under consideration by the NCI, Chemoprevention Branch. The NCI, Chemoprevention Branch is also considering a Phase II clinical efficacy trial of sulindac sulfone against breast and cervical neoplasia.

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Table I. Clinical Trials of Sulindac Sulfone Sponsored/Funded by NCI, DCPC

| Study No.<br>Title (PI)<br>Period of Performance<br>IND No. | Cancer Target | Study Population<br>No. of Subjects | Dose(s)<br>Treatment Duration | Endpoint(s) | Remarks |
|---|---|---|---|---|---|
| **Phase I (Safety and ADME)** | | | | | |
| NO1-CN-55119<br>Phase I Multiple-dose Safety and Pharmacokinetic Clinical Trial of Sulindac Sulfone (FGN-1) in Adenomatous Polyposis Coli Patients<br>(Dr. G. Thomas Budd,<br>The Cleveland Clinic Foundation)<br><br>6/95–12/96<br><br>IND 48,328 | --- | Adenomatous polyposis coli patients with subtotal colectomy<br><br>18 patients<br>6/arm | 200, 300 and 400 mg bid for 6 months | Multidose safety and ADME<br><br>Intermediate biomarkers: Apoptosis, polyp number/size, proliferation, loss of APC allele | Study is in progress; 15 patients have enrolled, 6 have completed the 200 mg bid dose and 2 have completed the 400 mg bid dose level. 21 patients have been screened to date. |
| **Phase II (Dose-titration, efficacy, and intermediate biomarkers)** | | | | | |
| Planned Study | Colon | Patients with previously resected colorectal adenomatous polyps (low-grade FAP cohort) | Oral dose selected from Phase I trial | Efficacy: New polyps and other intermediate biomarkers | Study is under consideration |
| Planned Study | Breast | Women with newly diagnosed DCIS | Oral dose selected from Phase I trial | Efficacy: Regression of DCIS, DNA ploidy, Proliferation, Apoptosis | Study is under consideration |
| Planned Study | Cervix | CIN II and III patients | Oral dose selected from Phase I trial | Efficacy: CIN regression, DNA ploidy, micronucleated cell frequency, *ras* expression, EGFR, keratins, involucrin | Study is under consideration |

## SULINDAC SULFONE DEVELOPMENT STATUS