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Design, Synthesis, and Pharmacological Evaluation of Glutamate Carboxypeptidase II (GCPII) Inhibitors Based on Thioalkylbenzoic Acid Scaffolds

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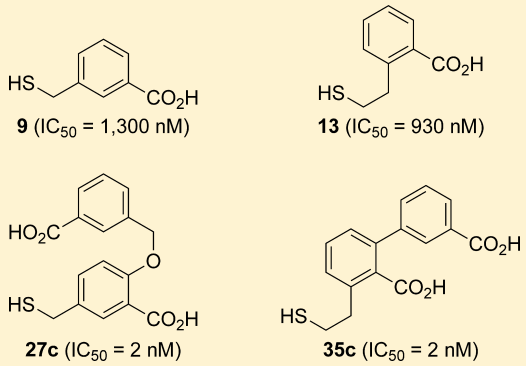
ABSTRACT: A series of thiol-based glutamate carboxypeptidase II (GCPII) inhibitors have been synthesized with either a 3-(mercaptomethyl)benzoic acid or 2-(2-mercaptoethyl)benzoic acid scaffold. Potent inhibitors were identified from each of the two scaffolds with IC_{50} values in the single-digit nanomolar range, including 2-(3-carboxybenzyloxy)-5-(mercaptomethyl)benzoic acid **27c** and 3-(2-mercaptoethyl)-biphenyl-2,3′-dicarboxylic acid **35c**. Compound **35c** was found to be metabolically stable and selective over a number of targets related to glutamate-mediated neurotransmission. Furthermore, compound **35c** was found to be orally available in rats and exhibited efficacy in an animal model of neuropathic pain following oral administration.



INTRODUCTION

One of the major sources of glutamate in the nervous system is believed to be from hydrolysis of the neuropeptide *N*-acetylaspartylglutamate (NAAG), catalyzed by a membrane-bound extracellular metalloprotease glutamate carboxypeptidase II (GCPII).[1] In theory, inhibition of GCPII would prevent the production of extracellular glutamate and, thereby, provide neuroprotective effects against excitotoxicity caused by excessive synaptic glutamate. Thus, substantial efforts have been made to identify potent and selective GCPII inhibitors, some of which have shown efficacy in a variety of animal models of neurological diseases.[2−4]

Nearly all potent GCPII inhibitors possess a zinc-binding group that interacts with the GCPII active site catalytic zinc atoms. Phosphonate,[5] phosphinate,[6] urea,[7,8] thiol,[9] and hydroxamate[10] were found to serve as an effective zinc binding group for GCPII inhibitors. Among these classes of GCPII inhibitors, those containing a thiol group offer a distinct pharmacological advantage over other series, particularly in chronic neurological disorders, because of their oral bioavailability. The first reported orally available GCPII inhibitor 2-(3-mercaptopropyl)pentanedioic acid **1** (2-MPPA, IC_{50} = 90 nM, Figure 1)[9] exhibited efficacy in various preclinical models of neurological disorders following oral administration including neuropathic pain,[9] amyotrophic lateral sclerosis (ALS),[11] diabetic neuropathy,[12] cocaine addiction,[13] and schizophrenia.[14] Compound **1** was subsequently tested in healthy subjects,

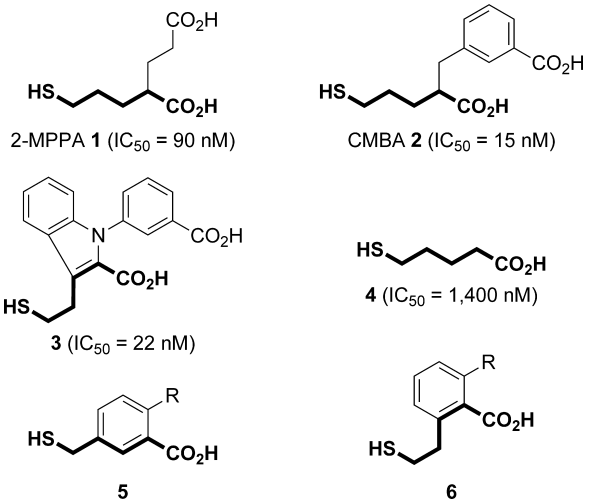


Figure 1. Thiol-based inhibitors of glutamate carboxypeptidase II (GCPII).

which represented the first administration of a GCPII inhibitor in humans.[15] Overall, **1** was safe and generally well-tolerated at plasma exposures that were effective in animal models of

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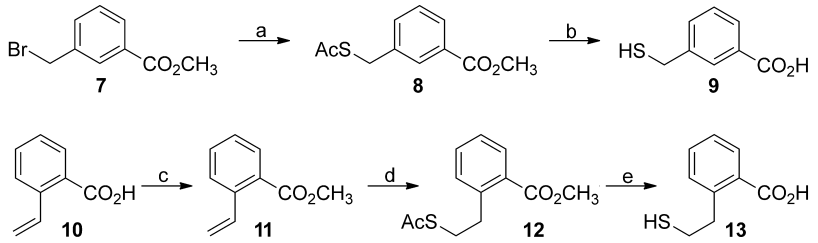
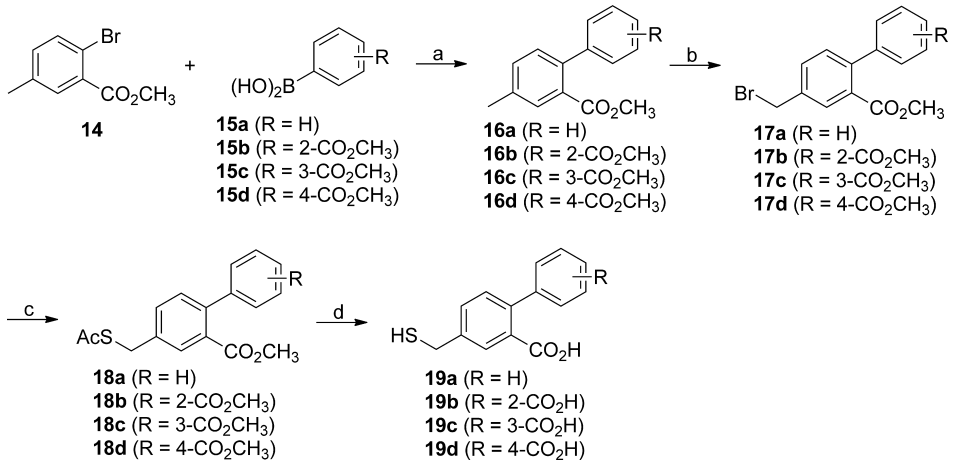
Scheme 1. Synthesis of 9 and 13[a]



[a]Reagents and conditions: (a) potassium thioacetate, acetone, 67 °C, 66%; (b) sodium hydroxide, water−dioxane, 100 °C, 46%; (c) methyl iodide, potassium carbonate, acetone, rt, 89%; (d) thioacetic acid, AIBN, toluene, 60 °C, 84%; (e) sodium hydroxide, water−dioxane, 100 °C, 96%.

Scheme 2. Synthesis of 19a−d[a]



[a]Reagents and conditions: (a) $Pd(PPh_3)_4$, potassium bicarbonate, DMF, 85 °C, 59−84%; (b) *N*-bromosuccinimide, AIBN, carbon tetrachloride, 77 °C, 78−100%; (c) potassium thioacetate, acetone, 67 °C, 23−71%; (d) sodium hydroxide, water−dioxane, reflux, 33−92%.

neuropathic pain. More importantly, no clinically significant adverse CNS effects normally associated with glutamate receptor antagonism were observed. The relatively higher oral doses (300 mg or higher) of **1** required for therapeutically relevant plasma concentrations, however, posed a risk for thiol-related adverse effects inducing immune reactions when conjugates are formed with endogenous proteins. Indeed, some of the adverse reactions reported for captopril are believed to be due in large part to its thiol group.[16]

Following the discovery of **1**, further efforts have been directed toward identifying new generations of thiol-based GCPII inhibitors with improved potency so that much lower doses can be sufficient for efficacy. For example, our group investigated the effect of P1′ side chain modifications on GCPII inhibitory potency using **1** as a template and identified 3-(2-carboxy-5-mercaptopentyl)benzoic acid **2** (CMBA, IC_{50} = 15 nM, Figure 1), bearing a 3-carboxybenzyl group at the P1′ side chain (Figure 1).[17] This represented the first successful attempt to improve the potency by modifying the glutarate moiety of the GCPII inhibitors. In a preclinical model of neuropathic pain, the effective dose (1.0 mg kg^{-1} day^{-1}) for **2** was found to be lower by an order of magnitude compared to that of **1** (10 mg kg^{-1} day^{-1}) presumably because of its improved potency toward GCPII. Interestingly, the benefit of introducing a 3-carboxybenzyl group to the P1′ side chain appears unique to the thiol-based GCPII inhibitors, as similar modifications to phosphonate-, phosphinate-, and hydroxamate-based GCPII inhibitors failed to enhance the inhibitory potency.[17] Further modification at the P1′ side chain resulted in the identification of GCPII inhibitors based on an indole scaffold.[18] 1-(3-Carboxyphenyl)-3-(2-mercaptoethyl)-1*H*-indole-2-carboxylic acid **3** potently inhibited GCPII with an IC_{50} of 22 nM (Figure 1). The common structural feature of all these thiol-based inhibitors is a 5-mercaptopentanoic acid backbone serving as the key pharmacophore for GCPII inhibition. Indeed, 5-mercaptopentanoic acid **4** itself is a low micromolar GCPII inhibitor (IC_{50} = 1400 nM) despite the absence of the P1′ side chain (Figure 1).[9] These findings prompted us to take advantage of the greater degree of structural diversity tolerated for thiol-based GCPII inhibitors and to further explore the possibility of incorporating other ring systems into the thiol-based pharmacophore.

Here we present a new class of thiol-based GCPII inhibitors **5** and **6** containing benzoic acid as a core ring system where the 5-mercaptopentanoic acid pharmacophore is embedded (Figure 1). Like the indole series, the new scaffolds possess a reduced number of rotatable bonds and increased lipophilicity in an attempt to achieve enhanced oral bioavailability. With an additional substituent at the ortho position of the benzoate moiety, a number of compounds were identified as potent GCPII inhibitors with IC_{50} values in the low nanomolar range. One of these compounds showed improved oral pharmacokinetic profiles in rats and exhibit in vivo efficacy superior to the existing thiol-based GCPII inhibitors in an animal model of neuropathic pain following oral administration.

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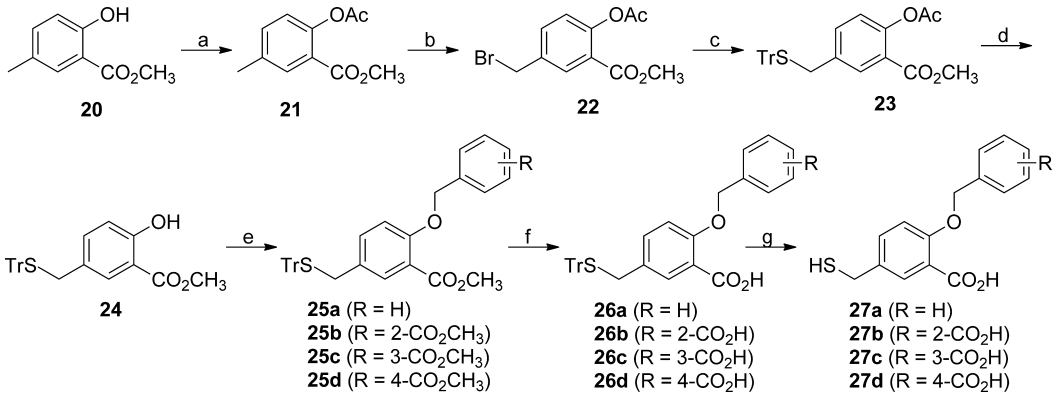
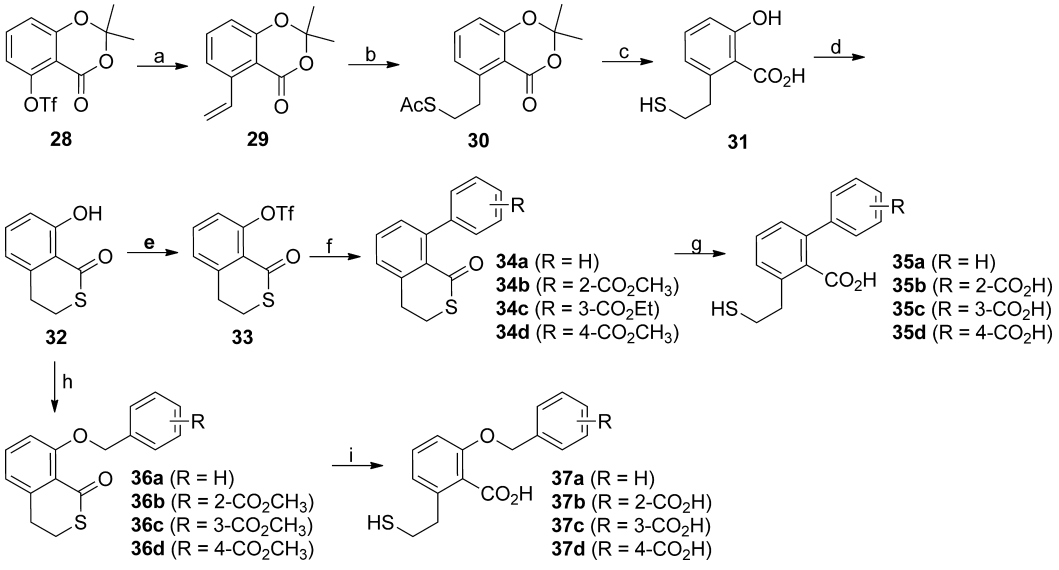
Scheme 3. Synthesis of 27a–d[a]



[a]Reagents and conditions: (a) Ac_2O, potassium carbonate, DMAP, dichloromethane, rt, 92%; (b) *N*-bromosuccinimide, AIBN, carbon tetrachloride, 77 °C, 84%; (c) TrSH, potassium carbonate, acetone, rt, 44%; (d) sodium methoxide, methanol, rt, 100% crude yield; (e) $RC_6H_4CH_2Br$, potassium carbonate, acetone, reflux, 57–70%; (f) sodium hydroxide, water–dioxane, reflux, 41–100%; (g) triisopropylsilane, TFA, dichloromethane, rt, 17–75%.

Scheme 4. Synthesis of 35a–d and 37a–d[a]



[a]Reagents and conditions: (a) $Pd(PPh_3)_4$, potassium carbonate, vinylboronic anhydride pyridine complex, 1,2-dimethoxyethane, 80 °C, 83%; (b) thioacetic acid, AIBN, toluene, 80 °C, 55%; (c) sodium hydroxide, TCEP, methanol–water, 65 °C, 93% crude yield; (d) *p*-TsOH, toluene, 110 °C, 71%; (e) Tf_2O, pyridine, dichloromethane, rt, 67%; (f) arylboronic acid, $Pd(PPh_3)_4$, cesium carbonate, toluene, 110 °C, 49–88%; (g) sodium hydroxide, water–dioxane, 110 °C, 53–68%; (h) $RC_6H_4CH_2Br$, potassium carbonate, acetone, 55 °C, 31–61%; (i) sodium hydroxide, water–dioxane, 100 °C, 13–61%.

RESULTS AND DISCUSSION

As shown in Scheme 1, compound **9** was synthesized from methyl 3-(bromomethyl)benzoate **7** by reacting with potassium thioacetate followed by base-mediated hydrolysis of both methyl ester and thioester. The synthesis of **13** started with methyl esterification of 2-vinylbenzoic acid **10** to **11** with methyl iodide. Radical-mediated addition of thioacetic acid to **11** gave the thioacetate **12**, and subsequent base-mediated hydrolysis afforded the desired compound **13**.

Scheme 2 illustrates the synthesis of compounds **19a–d**. Palladium-catalyzed Suzuki coupling of methyl 2-bromo-5-methylbenzoate **14** with phenylboronic acids **15a–d** provided biphenyl compounds **16a–d**. Bromination of **16a–d** with *N*-bromosuccinimide followed by the treatment with potassium thioacetate afforded **18a–d**. Subsequent base-mediated hydrolysis of **18a–d** gave the desired compounds **19a–d**.

Synthesis of **27a–d** is summarized in Scheme 3. Methyl 2-hydroxy-5-methylbenzoate **20** was converted into methyl 2-hydroxy-5-(tritylthiomethyl)benzoate **24** in four steps. This key intermediate was coupled with various benzyl bromides to form the 2-benzyloxybenzoates **25a–d**. Base-mediated hydrolysis of the methyl ester group(s) followed by reductive cleavage of the trityl group with triisopropylsilane/trifluoroacetic acid afforded the desired compounds **27a–d**.

As shown in Scheme 4, synthetic routes to **35a–d** and **37a–d** share the first four steps starting from the triflate **28**.[19] Suzuki coupling of **28** with vinylboronic anhydride catalyzed by tetrakis(triphenylphosphine)palladium[20] afforded the substituted styrene **29**. Radical-mediated addition of thioacetic acid to **29** gave the thioacetate **30**, and subsequent base-mediated hydrolysis provided compound **31**. Thiolactonization of **31** was carried out by heating the solution of **31** in toluene in the

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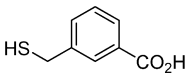
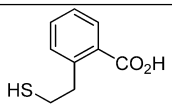
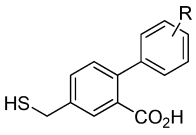
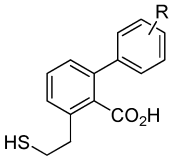
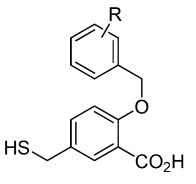
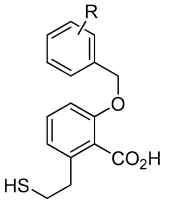
Table 1. Inhibition of GCPII by Thiol-Based Inhibitors[a]

Compd	Structure	IC_{50} (nM)	Compd	Structure	IC_{50} (nM)
9		1,300 ± 300	**13**		930 ± 630
19a (R = H)		5,700 ± 1,900	**35a** (R = H)		1,400 ± 300
19b (R = 2-CO_2H)		340 ± 120	**35b** (R = 2-CO_2H)		230 ± 140
19c (R = 3-CO_2H)		38 ± 28	**35c** (R = 3-CO_2H)		2 ± 1
19d (R = 4-CO_2H)		52 ± 13	**35d** (R = 4-CO_2H)		7 ± 4
27a (R = H)		420 ± 190	**37a** (R = H)		200 ± 60
27b (R = 2-CO_2H)		41 ± 16	**37b** (R = 2-CO_2H)		140 ± 30
27c (R = 3-CO_2H)		2 ± 1	**37c** (R = 3-CO_2H)		69 ± 6
27d (R = 4-CO_2H)		16 ± 8	**37d** (R = 4-CO_2H)		200 ± 150

[a]Values are the mean ± SD of three or more independent experiments.

presence of *p*-toluenesulfonic acid to give the common key intermediate **32**.

For the synthesis of **35a–d**, the intermediate **32** was first converted into the corresponding triflate **33** with triflic anhydride. Palladium-catalyzed Suzuki coupling of **33** with phenylboronic acids provided biphenyl compounds **34a–d**. Subsequent base-mediated hydrolysis of **34a–d** gave the desired compounds **35a–d**. Synthesis of **37a–d** involved Williamson reaction of **32** with various benzyl bromides to yield the benzyloxy derivatives **36a–d**. Subsequent base-mediated hydrolysis of **36a–d** gave the desired compounds **37a–d**.

Inhibitory potencies of the thiol-based compounds were measured using *N*-acetyl-L-aspartyl-[^{3}H]-L-glutamate as a substrate and purified human recombinant GCPII.[21] The results are summarized in Table 1. Both compounds **9** (IC_{50} = 1300 nM) and **13** (IC_{50} = 930 nM) exhibited inhibitory potencies comparable to 5-mercaptopentanoic acid **4** (IC_{50} = 1400 nM). Introduction of a phenyl ring into **9** resulted in nearly 5-fold reduction in potency as exemplified by compound **19a** with an IC_{50} of 5700 nM. Incorporation of a carboxyl group at any position on the phenyl ring, however, dramatically improved potency as in compounds **19b–d**. The significant improvement in potency by the addition of a carboxyl group suggests that these compounds take advantage of the positively charged residues located at the S1′ pocket of GCPII. Among them, the most potent compound **19c** inhibited GCPII activity with an IC_{50} of 38 nM comparable to that of **2** (IC_{50} = 15 nM) which shares its core skeletal frame.

Interestingly, benzyloxy-substituted derivatives **27a–d** exhibited GCPII inhibitory potency superior to that of their phenyl counterparts **19a–d**. As is the case with **19b–d**, increased potency was obtained by adding a carboxyl group into the phenyl ring of the benzyloxy moiety as in compounds **27b–d**. It is questionable, however, whether the bulky benzyloxy moiety of compounds **27a–d** occupies the S1′ site given the limited space available within the pocket. Although speculative, it is conceivable that the benzyloxy group of compounds **27a–d** is oriented toward the S1 pocket. Indeed, there are a few positively charged residues (Arg463, Arg534, and Arg536) at this site, which might explain the increased potency of compounds **27b–d** bearing a carboxyl group in the benzyloxy moiety over **27a**. A similar mode of "flipped" binding was proposed for sulfonamide-based GCPII inhibitors.[22] Among this series of GCPII inhibitors, 2-(3-carboxybenzyloxy)-5-(mercaptomethyl)benzoic acid **27c** was found to be a particularly potent GCPII inhibitor with an IC_{50} of 2 nM.

Introduction of a phenyl ring onto **13** led to an equally potent GCPII inhibitor **35a** with an IC_{50} of 1400 nM. In a fashion similar to compounds **19b–d**, addition of a carboxyl group to the phenyl ring provided more potent inhibitors **35b–d**. Among them, 2-(3-carboxyphenyl)-6-(2-sulfanylethyl)-benzoic acid **35c** exhibited extremely potent inhibition of GCPII with an IC_{50} of 2 nM. Like compound **19c**, the entire skeletal frame of **2** is embedded in compound **35c**. Nearly 10-fold improvement in potency of **35c** over **2** suggests that the rigid conformation provided by the biphenyl backbone of **35c** allows optimal interaction with GCPII. Benzyloxy-substituted derivative **37a** exhibited more potent GCPII inhibition than the parent compound **13**. Addition of a carboxyl group, however, did not significantly enhanced GCPII inhibitory potency in this series (compounds **37b–d**).

The overall SAR analysis indicates a reasonable correlation among the different scaffolds with respect to the effects of substitution pattern on GCPII inhibitory potency. In all cases, compounds containing a 3-carboxyphenyl group produced the most potent GCPII inhibitors as in compounds **19c**, **27c**, **35c**, and **37c**. In particular, compounds **27c** and **35c** represent the most potent thiol-based GCPII inhibitors reported to date. The two benzyloxy-substituted derivatives **27a** and **37a** exhibited IC_{50} values in the nanomolar range despite their lack of a second carboxyl group. While these compounds are not nearly as potent as **27c** and **35c**, their favorable druglike properties should warrant new SAR efforts strictly on monocarboxylate compounds within and beyond the benzyloxy substituted derivatives.

Further pharmacological characterization was conducted with one of the most potent GCPII inhibitors, compound **35c**. No

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evidence of mutagenicity was observed in the Ames test using *Salmonella typhimurium* strains (TA98, TA100, TA1535, and TA1537) in the presence or absence of metabolic activation by S9 mixture. Compound **35c** was also tested in a panel of in vitro assays including those related to glutamate-mediated neurotransmission. As shown in Table 2, compound **35c** showed no significant activity in any of these assays at 10 μM (>1000-fold greater than IC_{50} of **35c**) or higher concentration. In vitro metabolic stability of **35c** was examined by using liver microsomes of mouse, rat, dog, monkey, and human. No significant metabolism of **35c** was detected across all species during a 2 h incubation period. In vivo pharmacokinetic studies of **35c** were carried out in fasted rats by intravenous and oral administration (10 mg/kg). The plasma pharmacokinetic parameters are summarized in Table 3. The oral bioavailability of **35c** was approximately 40%. Absorption of **35c** was relatively rapid and the plasma peak concentration was generally observed within the first hour with a terminal half-life of 3−4 h. It is worth noting that the AUC followed by 10 mg/kg oral administration of **35c** (33.8 μg·h/mL) is nearly 20-fold higher than that of **1** (1.82 μg·h/mL)[9] as a result of the higher C_{max} and lower clearance.

Table 2. In Vitro Pharmacological Characterization of 35c

assay	results
hERG channel	no inhibition up to 100 μM
NMDA receptor (agonism)	<5% inhibition at 10 μM
NMDA receptor (glycine)	12% inhibition at 10 μM
NMDA receptor (phencyclidine)	no inhibition at 10 μM
NMDA receptor (polyamine)	no inhibition at 10 μM
AMPA receptor	no inhibition at 10 μM
kainate receptor	no inhibition at 10 μM
glycine receptor (strychnine sensitive)	no inhibition at 10 μM
glutaminase (kidney-type)	no inhibition up to 100 μM

Compound **35c** was subsequently tested for its antinociceptive effects following oral administration (0.1 mg kg^{-1} day^{-1}) using the rat chronic constriction injury model of neuropathic pain.[23] As shown in Figure 2, **35c** significantly reduced thermal hyperalgesia relative to the vehicle-treated control on days 8 and 12. The effective dose for **35c** is lower by 2 orders of magnitude compared to that of **1** (10 mg kg^{-1} day^{-1})[9] and by 1 order of magnitude compared to that of **2** (1.0 mg kg^{-1} day^{-1}),[17] presumably because of its enhanced GCPII inhibitory potency coupled with the improved pharmacokinetic properties.

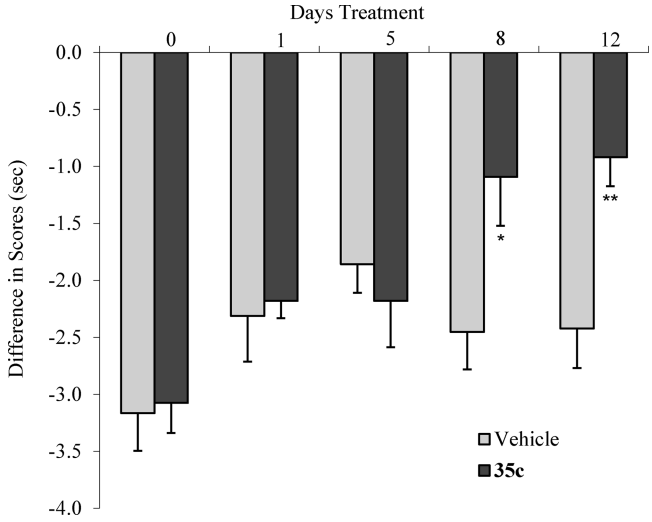

Figure 2. Antinociceptive effects of compound **35c** in the rat chronic constriction injury (CCI) model of neuropathic pain. Oral administration of **35c** at 0.1 mg kg^{-1} day^{-1} significantly attenuated CCI-induced hyperalgesic state relative to the vehicle-treated control (*, $p < 0.05$; **, $p < 0.005$).

CONCLUSIONS

Unlike other zinc-binding groups, the thiol group is nucleophilic and prone to oxidation. These chemical properties are linked to the risk of inducing immune reactions when conjugates are formed with endogenous proteins. Indeed, some of the adverse reactions reported for captopril are believed to be due in large part to its thiol group.[16] Since more serious reactions can be managed by decreasing doses, as in the case of captopril, it is vital to identify highly potent thiol-based GCPII inhibitors with low efficacious doses in order to minimize the thiol-related side effects. We synthesized a series of compounds based on either 3-(mercaptomethyl)benzoic acid **9** or 2-(2-mercaptoethyl)benzoic acid **13** scaffold. Some of these compounds were found to be very potent inhibitors of GCPII. 2-(3-Carboxyphenyl)-6-(2-sulfanylethyl)benzoic acid **35c** was found to be orally available and efficacious in an animal model of peripheral neuropathy by oral administration. Given the low dose (0.1 mg/kg) required for efficacy, compound **35c** and other potent inhibitors reported herein may represent a new class of GCPII inhibitors with significant therapeutic potential. Further dose−response pharmacological evaluations of **35c** are currently underway in various animal models of neurological disorders associated with glutamate excitotoxicity.

Furthermore, the synthetic routes developed herein will allow us to utilize widely available arylboronic acids and benzyl bromides, leading to more extensive SAR studies on derivatives based on scaffolds **9** and **13**. In addition, very potent GCPII inhibitors emerging from present and future SAR efforts should serve as valuable templates into which other zinc-binding groups can be incorporated to further expand the structural diversity of GCPII inhibitors.

EXPERIMENTAL SECTION

General. All solvents were reagent grade or HPLC grade. Unless otherwise noted, all materials were obtained from commercial suppliers and used without further purification. All reactions were performed under nitrogen. Preparative HPLC purification was performed on an Agilent 1200 series HPLC system equipped with a multiwavelength detector using a Phenomenex Luna 5 μm C18 (2) column (250 mm × 4.6 mm) with a flow rate of 15 mL/min. The

Table 3. Pharmacokinetic Profile of 35c in Fasted Rats

dose	C_{max} (μg/mL)	T_{max} (h)	AUC_{inf} (μg·h/mL)	CL/F (mL h^{-1} kg^{-1})	$T_{1/2}$ (h)	F (%)
10 mg/kg iv (n = 4)			90.6 ± 9.0	267 ± 120	3.6 ± 0.6	
10 mg/kg po (n = 9)	7.10 ± 5.55	1.1 ± 0.6	33.8 ± 9.9	318 ± 90	3.2 ± 0.6	37

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solvent system consisted of distilled water with 0.1% formic acid (solvent A) and acetonitrile with 0.1% formic acid (solvent B). 40% B was used from 0 to 5 min followed by a linear gradient from 40% to 85% B over 35 min. Melting points were obtained on a Mel-Temp apparatus and are uncorrected. ^{1}H NMR spectra were recorded at 400 MHz. ^{13}C NMR spectra were recorded at 100 MHz. Elemental analyses were performed by Atlantic Microlab (Norcross, GA) and were within ±0.4% of calculated values.

Methyl 3-(Acetylsulfanylmethyl)benzoate (8). To a solution of methyl 3-(bromomethyl)benzoate **7** (1.00 g, 4.37 mmol) in acetone (60 mL) was added potassium thioacetate (0.60 g, 5.24 mmol), and the resulting mixture was stirred at 67 °C for 1.5 h. The mixture was filtered, and the filtrate was concentrated in vacuo. The residue was purified by silica gel chromatography (9:1 hexanes/EtOAc) to provide 0.64 g of **8** as clear oil (66% yield): ^{1}H NMR (DMSO-d_6) δ 2.36 (s, 3H), 3.85 (s, 3H), 4.19 (s, 2H), 7.47 (t, J = 7.6 Hz, 1H), 7.57 (m, 1H), 7.83 (d, J = 7.8 Hz, 1H), 7.90 (m, 1H).

3-(Sulfanylmethyl)benzoic Acid (9). To a solution of **8** (0.30 g, 1.34 mmol) in H_2O−dioxane (1:1, 6 mL) was added sodium hydroxide (0.21 g, 5.25 mmol, 4.0 equiv). The resulting mixture was heated at 100 °C for 1 h. The mixture was cooled to room temperature, and one spatula tip of tris(2-carboxyethyl)phosphine was added. After the mixture was stirred for 15 min, the solvents were removed in vacuo. The resulting residue was acidified with aqueous 10% $KHSO_4$ and the product extracted with ethyl acetate. The organic layer was washed with brine, dried over $MgSO_4$, and concentrated in vacuo. The resulting solid was purified by reverse-phase HPLC to provide 0.103 g of **9** as a white crystal (46% yield): mp 104−106 °C; ^{1}H NMR (DMSO-d_6) δ 3.00 (t, J = 8.0 Hz, 1H), 3.81 (d, J = 7.8 Hz, 2H), 7.45 (t, J = 7.6 Hz, 1H), 7.59 (d, J = 7.1 Hz, 1H), 7.80 (d, J = 8.1 Hz, 1H), 7.94 (s, 1H), 12.99 (s, 1H). Anal. Calcd for $C_8H_8O_2S$: C, 57.12; H, 4.79; S, 19.06. Found: C, 57.27; H, 4.98; S, 18.94.

Methyl 2-Vinylbenzoate (11). To a solution of 2-vinylbenzoic acid **10** (0.16 g, 1.08 mmol) in acetone (10 mL) was added potassium carbonate (0.30 g, 2.16 mmol). The mixture was allowed to stir for 5 min, after which methyl iodide (0.10 mL, 1.62 mmol, 1.5 equiv) was added. After 24 h at room temperature, the solvent was removed in vacuo and the residue was taken up by ethyl acetate and washed with water and brine. The organic layer was dried over sodium sulfate and concentrated to yield 0.156 g of **11** as a colorless oil (89% crude yield). The crude material was used as is without purification: ^{1}H NMR ($CDCl_3$) δ 3.92 (s, 3H), 5.37 (dd, J = 1.3, 10.9 Hz, 1H), 5.64 (dd, J = 1.3,17.4 Hz, 1H), 7.32 (dt, J = 1.3, 7.6 Hz, 1H), 7.43 (d, J = 11.1 Hz, 1H), 7.48−7.53 (m, 1H), 7.58 (m, 1H), 7.89 (dt, J = 0.8, 7.8 Hz, 1H).

Methyl 2-(2-(Acetylthio)ethyl)benzoate (12). To a solution of **11** (0.05 g, 0.31 mmol) in toluene (0.5 mL) was added thioacetic acid (0.44 mL, 6.16 mmol). The mixture was heated at 60 °C, after which AIBN (0.005 g, 0.031 mmol) was added in one portion. After 1 h at 60 °C, the mixture was cooled to room temperature. Solvent was removed under reduced pressure, and excess thioacetic acid was removed by coevaporation with toluene (25 mL × 2). The crude product was purified by a Biotage flash chromatography apparatus (SNAP 10 g cartridge) using 5−40% gradient of ethyl acetate in hexanes to yield 0.061 g of **12** as a colorless oil (84% yield): ^{1}H NMR ($CDCl_3$) δ 2.34 (s, 2H), 3.17 (m, 2H), 3.23 (m, 2H), 3.92 (s, 3H), 7.31−7.34 (m, 2H), 7.47 (dt, J = 1.56, 7.6 Hz, 1H), 7.93 (dd, J = 1.1, 7.7 Hz, 1H); ^{13}C NMR ($CDCl_3$) δ 30.2, 30.6, 34.5, 52.1, 126.7, 129.3, 130.9, 131.6, 132.2, 141.6, 167.7, 195.8.

2-(2-Mercaptoethyl)benzoic Acid (13). To a solution of **12** (0.047 g, 0.20 mmol) in water−dioxane mixture (1:1, 0.5 mL) was added sodium hydroxide (0.035 g, 0.88 mmol) in one portion. The reaction mixture was heated at 100 °C for 1 h, followed by the addition of one spatula tip of TCEP. After the mixture was stirred for an additional 20 min at room temperature, solvents were removed in vacuo and the residue was taken up by ethyl acetate and water. The aqueous layer was then acidified to pH ∼4 with aqueous 10% $KHSO_4$ and extracted with ethyl acetate three times. The combined organic extracts were washed with brine, dried over sodium sulfate, and concentrated to give 0.035 g of **13** as an off-white solid (96% yield): mp 84−85 °C; ^{1}H NMR ($CDCl_3$) δ 1.45 (t, J = 8.1 Hz, 1H), 2.85 (dd, J = 7.3, 15.2 Hz, 2H), 3.36 (t, J = 7.8 Hz, 2H), 7.35 (m, 2H), 7.54 (m, 1H), 8.12 (dd, J = 1.3, 7.8 Hz, 1H); ^{13}C NMR ($CDCl_3$) δ 25.9, 39.3, 126.8, 127.9, 131.9, 132.0, 133.2, 142.7, 172.8; MS ES$^-$ m/z 181.1 (M − H) $^-$; MS ES$^+$ m/z 183.0 (M + 1)$^+$. Anal. Calcd for $C_9H_{10}O_2S$: C, 59.32; H, 5.53; S, 17.60. Found: C, 59.02; H, 5.71; S, 17.30.

Methyl 5-Methyl-2-phenylbenzoate (16a). To a solution of methyl 2-bromo-5-methylbenzoate **14** (1.00 g, 4.37 mmol) in DMF (10 mL) were added phenylboronic acid **15a** (0.64 g, 5.25 mmol), potassium bicarbonate (1.99 g, 19.9 mmol), and tetrakis-(triphenylphosphine)palladium (0.15 g, 0.13 mmol). The mixture was heated at 85 °C for 3.5 h. The solvent was removed in vacuo, and the residue was taken up by ethyl acetate (40 mL). The solution was subsequently washed with water, aqueous 10% $KHSO_4$, and brine. The solution was then dried over $MgSO_4$ and concentrated. The residue was purified by silica gel chromatography (5% EtOAc in hexanes) to provide 0.83 g of **16a** as a clear oil (84% yield): ^{1}H NMR (DMSO-d_6) δ 2.38 (s, 3H), 3.57 (s, 3H), 7.25 (m, 2H), 7.33 (m, 2H), 7.39−7.44 (m, 3H), 7.54 (s, 1H).

Methyl 2-(2-Methoxycarbonylphenyl)-5-methylbenzoate (16b). Compound **16b** was prepared as described for the preparation of **16a** except (2-methoxycarbonylphenyl)boronic acid **15b** was used in place of phenylboronic acid **15a**: clear oil (59% yield); ^{1}H NMR (DMSO-d_6) δ 2.40 (s, 3H), 3.51 (s, 3H), 3.52 (s, 3H), 7.11 (d, J = 7.8 Hz, 1H), 7.19−7.24 (m, 1H), 7.41 (m, 1H), 7.51 (m, 1H), 7.59 (m, 1H), 7.69 (s, 1H), 7.84−7.89 (m, 1H).

4-Methyl-[1,1′-biphenyl]-2,3′-dicarboxylic Acid Dimethyl Ester (16c). Compound **16c** was prepared as described for the preparation of **16a** except (3-methoxycarbonylphenyl)boronic acid **15c** was used in place of phenylboronic acid **15a**: colorless oil (82% yield); ^{1}H NMR ($CDCl_3$) δ 2.44 (s, 3H), 3.64 (s, 3H), 3.93 (s, 3H), 7.28 (t, J = 3.9 Hz, 1H), 7.36 (m, 1H), 7.46−7.49 (m, 2H), 7.71 (m, 1H), 7.99−8.04 (m, 2H); ^{13}C NMR ($CDCl_3$) δ 21.0, 51.9, 52.1, 127.9, 128.2, 129.5, 130.6, 130.7, 131.5, 132.3, 133.0, 137.6, 138.8, 140.4, 141.6, 167.0, 168.7.

Methyl 2-(4-Methoxycarbonylphenyl)-5-methylbenzoate (16d). Compound **16d** was prepared as described for the preparation of **16a** except (4-methoxycarbonylphenyl)boronic acid **15d** was used in place of phenylboronic acid **15a**: clear oil (65% yield); ^{1}H NMR (DMSO-d_6) δ 2.39 (s, 3H), 3.58 (s, 3H), 3.87 (s, 3H), 7.33 (d, J = 7.8 Hz, 1H), 7.39 (d, J = 8.6 Hz, 2H), 7.44 (m, 1H), 7.61 (m, 1H), 7.97 (d, J = 8.3 Hz, 2H).

Methyl 5-(Bromomethyl)-2-phenylbenzoate (17a). To a solution of **16a** (0.81 g, 3.58 mmol) in CCl_4 (10 mL) were added *N*-bromosuccinimide (0.70 g, 3.93 mmol) and AIBN (0.12 g, 0.73 mmol). The mixture was heated at 77 °C for 3.5 h and then cooled to room temperature. The white precipitate was filtered off and the filtrate was concentrated in vacuo to give 0.85 g of **17a** as a brown oil (78% crude yield). The crude material was used for the next reaction without further purification: ^{1}H NMR (DMSO-d$_6$) δ 3.60 (s, 3H), 4.82 (s, 2H), 7.28 (m, 2H), 7.36−7.46 (m, 4H), 7.68 (dd, J = 2.0, 7.8 Hz, 1H), 7.82 (d, J = 1.8 Hz, 1H).

Dimethyl 4-(Bromomethyl)biphenyl-2,2′-dicarboxylate (17b). Compound **17b** was prepared as described for the preparation of **17a** except methyl 2-(2-methoxycarbonylphenyl)-5-methylbenzoate **16b** was used in place of **16a**: brown oil (100% crude yield); ^{1}H NMR (DMSO-d_6) δ 3.54 (s, 6H), 4.83 (s, 2H), 7.21 (m, 2H), 7.49 (t, J = 7.6 Hz, 1H), 7.61 (t, J =, 1H), 7.66 (d, J = 7.8 Hz, 1H), 7.89 (d, J = 7.8 Hz, 1H), 7.96 (s, 1H).

Dimethyl 4-(Bromomethyl)biphenyl-2,3′-dicarboxylate (17c). Compound **17c** was prepared as described for the preparation of **17a** except methyl 2-(3-methoxycarbonylphenyl)-5-methylbenzoate **16c** and benzoyl peroxide were used in place of **16a** and AIBN, respectively: brown oil (60% yield); ^{1}H NMR (DMSO-d_6) δ 3.61(s, 3H), 3.88 (s, 3H), 4.84 (s, 2H), 7.48−7.50 (d, J = 8.0 Hz, 1H), 7.59−7.60 (m, 2H), 7.72−7.75 (m, 1H), 7.85 (s, 1H), 7.90 (m, 1H), 7.97−7.99 (m, 1H).

Dimethyl 4-(Bromomethyl)biphenyl-2,4′-dicarboxylate (17d). Compound **17d** was prepared as described for the preparation of **17a** except methyl 2-(4-methoxycarbonylphenyl)-5-methylbenzoate **16d** was used in place of **16a**: brown oil (94% crude yield); ^{1}H NMR

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(DMSO-d_6) δ 3.60 (s, 3H), 3.88 (s, 3H), 4.83 (s, 2H), 7.43 (d, J = 8.1 Hz, 2H), 7.46 (d, J = 7.8 Hz, 1H), 7.72 (dd, J = 1.8, 7.8 Hz, 1H), 7.90 (m, 1H), 7.99 (d, J = 8.1 Hz, 2H).

Methyl 5-(Acetylsulfanylmethyl)-2-phenylbenzoate (18a). To a solution of **17a** (0.59 g, 1.93 mmol) in acetone (30 mL) was added potassium thioacetate (0.27 g, 2.36 mmol), and the mixture was heated at 67 °C for 1.5 h. The reaction mixture was cooled to room temperature, and excess potassium carbonate was filtered. The filtrate was concentrated in vacuo. The residue was purified by reverse-phase HPLC (method, 40−100% acetonitrile−water, 0.1% formic acid) to give 0.41 g of **18a** as white powder (71% yield): ^{1}H NMR (DMSO-d_6) δ 2.37 (s, 3H), 3.57 (s, 3H), 4.20 (s, 2H), 7.25 (m, 2H), 7.35−7.44 (m, 4H), 7.52 (dd, J = 2.0, 8.1 Hz, 1H), 7.65 (d, J = 1.8 Hz, 1H).

Methyl 5-(Acetylsulfanylmethyl)-2-(2-methoxycarbonylphenyl)benzoate (18b). Compound **18b** was prepared as described for the preparation of **18a** except dimethyl 4-(bromomethyl)biphenyl-2,2′-dicarboxylate **17b** was used in place of **17a**: white powder (23% yield); ^{1}H NMR (DMSO-d_6) δ 2.40 (s, 3H), 3.52 (s, 3H), 3.53 (s, 3H), 4.23 (s, 2H), 7.15 (d, J = 7.8 Hz, 1H), 7.21 (dd, J = 1.3, 8.1 Hz, 1H), 7.48 (dt, J = 1.5, 7.6, 1H), 7.50 (dd, J = 1.8, 7.8 Hz, 1H), 7.60 (dt, J = 1.3, 7.3 Hz, 1H), 7.81 (d, J = 2.02 Hz, 1H), 7.87 (dd, J = 1.5, 7.8 Hz, 1H).

Dimethyl 4-(Acetylthiomethyl)biphenyl-2,3′-dicarboxylate (18c). Compound **18c** was prepared as described for the preparation of **18a** except dimethyl 4-(bromomethyl)biphenyl-2,3′-dicarboxylate **17c** was used in place of **17a** (76% yield): ^{1}H NMR (DMSO-d_6) δ 2.39 (s, 3H), 3.60 (s, 3H), 3.88 (s, 3H), 4.23 (s, 2H), 7.42−7.44 (d, J = 8.0 Hz, 1H), 7.57−7.60 (m, 3H), 7.74 (s 1H), 7.83 (s, 1H), 7.96−7.99 (m, 1H).

Dimethyl 4-(Acetylthiomethyl)biphenyl-2,4′-dicarboxylate (18d). Compound **18d** was prepared as described for the preparation of **18a** except dimethyl 4-(bromomethyl)biphenyl-2,4′-dicarboxylate **17d** was used in place of **17a** (33% yield): ^{1}H NMR (DMSO-d_6) δ 2.39 (s, 3H), 3.59 (s, 3H), 3.88 (s, 3H), 4.22 (s, 2H), 7.42 (d, J = 8.1 Hz, 3H), 7.58 (dd, J_1 = 2.0, 8.1 Hz, 1H), 7.74 (d, J = 2.0 Hz, 1H), 7.99 (d, J = 8.6 Hz, 2H).

4-(Mercaptomethyl)biphenyl-2-carboxylic Acid (19a). To a solution of **18a** (0.41 g, 1.36 mmol) in H_2O/dioxane (6 mL, 1:1) was added sodium hydroxide (0.22 g, 5.50 mmol), and the resulting mixture was stirred at 100 °C for 1 h. The reaction mixture was cooled to room temperature, and one spatula tip of tris(2-carboxyethyl)-phosphine was added. After the mixture was stirred for an additional 15 min, the solvents were removed in vacuo. The resulting residue was acidified with aqueous 10% $KHSO_4$ and extracted with ethyl acetate. The organic layer was washed with brine, dried over $MgSO_4$, and concentrated. The resulting solid was recrystallized in EtOAc/hexanes to give 0.11 g of **19a** as a white solid (33% yield): mp 179−181 °C; ^{1}H NMR (DMSO-d_6) δ 3.03 (t, J = 7.8 Hz, 1H), 3.81 (d, J = 7.8 Hz, 2H), 7.30−7.34 (m, 3H), 7.35−7.42 (m, 3H), 7.52 (m, 1H), 7.69 (d, J = 1.8 Hz, 1H), 12.77 (bs, 1H); ^{13}C NMR (DMSO-d_6) δ 27.01, 127.1, 127.2, 128.1, 128.26, 128.29, 128.8, 130.0, 130.5, 130.6, 139.2, 140.6, 140.9, 169.6. Anal. Calcd. for $C_{14}H_{12}O_2S$: C, 68.83; H, 4.95; S, 13.12. Found: C, 68.56; H, 5.16; S, 12.85.

4-(Mercaptomethyl)biphenyl-2,2′-dicarboxylic Acid (19b). Compound **19b** was prepared as described for the preparation of **19a** except methyl 5-(acetylsulfanylmethyl)-2-(2-methoxycarbonylphenyl)benzoate **18b** was used in place of **18a**: white powder (38% yield): mp 203−205 °C; ^{1}H NMR (DMSO-d_6) δ 3.03 (t, J = 8.1 Hz, 1H), 3.82 (d, J = 8.1 Hz, 2H), 7.09 (d, J = 7.8 Hz, 2H), 7.13 (dd, J = 1.1, 7.6 Hz, 1H), 7.42 (dt, J = 1.5, 7.6 Hz, 1H), 7.51 (m, 2H), 7.86 (m, 2H), 12.46 (s, 2H); ^{13}C NMR (DMSO-d_6) δ 27.1, 126.9, 129.3, 129.5, 130.4, 130.4, 130.4, 130.5, 130.8, 131.0, 140.3, 141.4, 142.8, 167.7, 167.9. Anal. Calcd. for $C_{15}H_{12}O_4S$·0.36EtOAc: C, 61.70; H, 4.69; S, 10.02. Found: C, 62.07; H, 4.54; S, 9.64.

4-(Mercaptomethyl)biphenyl-2,3′-dicarboxylic Acid (19c). Compound **19c** was prepared as described for the preparation of **19a** except dimethyl 4-(acetylthiomethyl)biphenyl-2,3′-dicarboxylate **18c** was used in place of **18a**. A silica gel column chromatography (dichloromethane/EtOAc, 9:1 with 1% acetic acid) was used instead of recrystallization for the purification of the crude material: white solid (92% yield); ^{1}H NMR (DMSO-d_6) δ 3.10 (t, J = 8.0 Hz, 1H), 3.89 (d, J = 8.0 Hz, 2H), 7.43 (d, J = 7.5 Hz, 1H), 7.58−7.65 (m, 3H), 7.83 (d, J = 2.0 Hz, 1H), 7.92 (s, 1H), 7.97−8.00 (m, 1H). Anal. Calcd for $C_{15}H_{12}O_4S$·0.5AcOH: C, 60.37; H, 4.43; O, 25.13; S, 10.07. Found: C, 60.28; H, 4.45; S, 10.15.

4-(Mercaptomethyl)biphenyl-2,4′-dicarboxylic Acid (19d). Compound **19d** was prepared as described for the preparation of **19a** except dimethyl 4-(acetylthiomethyl)biphenyl-2,4′-dicarboxylate **18d** was used in place of **18a**: white powder (53% yield): mp 236−238 °C; ^{1}H NMR (DMSO-d_6) δ 3.06 (t, J = 8.1 Hz, 1H), 3.83 (d, J = 8.1 Hz, 2H), 7.36 (d, J = 7.8 Hz, 1H), 7.43 (d, J = 8.6 Hz, 2H), 7.57 (dd, J = 2.0, 7.8 Hz, 1H), 7.77 (d, J = 2.0 Hz, 1H), 7.94 (d, J = 8.1 Hz, 2H), 12.92 (bs, 2H). Anal. Calcd. for $C_{15}H_{12}O_4S$·0.4EtOAc: C, 61.62; H, 4.73; S, 9.91. Found: C, 62.02; H, 4.37; S, 9.55.

Methyl 2-Acetoxy-5-methylbenzoate (21). To a solution of methyl 2-hydroxy-5-methylbenzoate **20** (10 mL, 70 mmol), potassium carbonate (14.5 g, 105 mmol), and DMAP (50 mg, 0.41 mmol) in dichloromethane (50 mL) was dropwise added acetic anhydride (7.2 mL, 77 mmol). The reaction mixture was stirred for 72 h at room temperature. The reaction was quenched by the addition of 300 mL of 10% $KHSO_4$. The aqueous phase was extracted with dichloromethane twice (300 and 150 mL). The combined organic phases were dried over $MgSO_4$, filtered, and concentrated in vacuo to afford 13.3 g of **21** as a light yellow oil (92% crude yield). This material was used without further purification: ^{1}H NMR ($CDCl_3$) δ 2.34 (s, 3H), 2.37 (s, 3H), 3.86 (s, 3H), 6.98 (d, J = 8.3 Hz, 1H), 7.35 (m, 1H), 7.82 (m, 1H).

Methyl 2-Acetoxy-5-(bromomethyl)benzoate (22). To a solution of **21** (9.13 g, 43.9 mmol) in CCl_4 (65 mL) were added *N*-bromosuccinimide (9.77 g, 54.9 mmol) and AIBN (1.3 g, 7.9 mmol). The reaction mixture was heated at 77 °C for 4 h. The reaction mixture was poured into 10% $KHSO_4$ (60 mL) and extracted with dichloromethane (60 mL × 2). The combined organic extracts were dried over $MgSO_4$, filtered, and concentrated. The crude material was purified by a silica gel chromatography (10% ethyl acetate/hexanes) to afford 10.6 g of **22** as a white solid (84% yield): ^{1}H NMR ($CDCl_3$) δ 2.35 (s, 3H), 3.88 (s, 3H), 4.48 (s, 2H), 7.09 (d, J = 8 Hz, 1H), 7.58 (dd, J = 8.4, 2.3 Hz, 1H), 8.04 (d, J = 2.0 Hz, 1H).

Methyl 2-Acetoxy-5-(tritylthiomethyl)benzoate (23). To a solution of **22** (5.52 g, 19.2 mmol) in acetone (30 mL) was added potassium carbonate (4.0 g, 28.5 mmol) followed by triphenylmethanethiol (5.76 g, 20.9 mmol). The mixture was stirred for 14 h at room temperature. The excess potassium carbonate was removed by filtration, and the filtrate was concentrated in vacuo. The resultant residue was reconstituted in EtOAc (100 mL). The organic layer was washed with 10% $KHSO_4$ (75 mL). The aqueous phase was extracted again with EtOAc (100 mL). The combined organic extracts were dried over $MgSO_4$, filtered, and concentrated in vacuo. The crude material was purified by trituration with a mixture of MeOH and EtOAc to give 4.07 g of **23** as a light yellow crystal (44% yield): ^{1}H NMR ($CDCl_3$) δ 2.32 (s, 3H), 3.35 (s, 2H), 3.85 (s, 3H), 6.96 (d, J = 8.4 Hz, 1H), 7.21−7.32 (m, 10H), 7.43−7.46 (m, 6H), 7.72 (d, J = 2.3 Hz, 1H).

Methyl 2-Hydroxy-5-(tritylthiomethyl)benzoate (24). To a solution of **23**, (4.07 g, 8.4 mmol) in MeOH (80 mL) was added sodium methoxide (684 mg, 12.7 mmol) portionwise at 0 °C. The mixture was warmed to room temperature overnight. The reaction was quenched with 10% $KHSO_4$ (30 mL), and then the mixture was concentrated to dryness. The residue was dissolved in EtOAc, and the organic layer was washed with 10% $KHSO_4$. The organic layer was dried over $MgSO_4$, filtered, and concentrated to give 3.62 g of **24** (100% crude yield). The crude material was used in the subsequent step without further purification: ^{1}H NMR ($CDCl_3$) δ 3.27 (s, 2H), 3.94 (s, 3H), 6.86 (d, J = 8.5 Hz, 1H), 7.19−7.25 (m, 4H), 7.28−7.33 (m, 6H), 7.44−7.47 (m, 6H), 7.58 (d, J = 2.3 Hz, 1H), 10.67 (s, 1H).

Methyl 2-(Benzyloxy)-5-(tritylthiomethyl)benzoate (25a). To a solution of **24** (0.500 g, 1.13 mmol) in acetone (10 mL) was added potassium carbonate (0.188 g, 1.36 mmol) followed by benzyl bromide (0.233 g, 1.36 mmol). The mixture was heated at reflux for 72 h. The excess potassium carbonate was removed by filtration. The filtrate was concentrated in vacuo, and the residue was partitioned

between EtOAc (50 mL) and 10% $KHSO_4$ (50 mL). The organic layer was separated, and the aqueous layer was extracted with EtOAc (50 mL). The combined organic extracts were dried over $MgSO_4$, filtered, and concentrated. The crude material was purified by a silica gel column chromatography (20% EtOAc in hexanes) to give 0.320 g of **25a** as a white solid (53% yield): ^{1}H NMR (DMSO-d_6) δ 3.28 (s, 2H) 3.80 (s, 3H) 5.18 (s, 2H) 7.02−7.54 (m, 23H).

Methyl 2-(2-(Methoxycarbonyl)benzyloxy)-5-(tritylthiomethyl)benzoate (25b). Compound **25b** was prepared as described for the preparation of **25a** except methyl 2-(bromomethyl)benzoate was used in place of benzyl bromide: white solid (66% yield); ^{1}H NMR ($CDCl_3$) δ 3.29 (s, 2H), 3.91 (s, 3H), 3.93 (s, 3H), 5.55 (s, 2H), 6.96 (d, J = 8.0 Hz, 1H), 7.18−7.25 (m, 4H), 7.28−7.32 (m, 6H), 7.38 (dt, J = 8.1, 1.6 Hz, 1H), 7.46 (m, 6H), 7.59−7.63 (m, 2H), 8.04 (dd, J = 8.0, 2.1 Hz, 2H).

Methyl 2-(3-(Methoxycarbonyl)benzyloxy)-5-(tritylthiomethyl)benzoate (25c). Compound **25c** was prepared as described for the preparation of **25a** except methyl 3-(bromomethyl)benzoate was used in place of benzyl bromide: white solid (70% yield); ^{1}H NMR ($CDCl_3$) δ 3.28 (s, 2H), 3.92 (s, 3H), 3.93 (s, 3H), 5.17 (s, 2H), 6.87 (d, J = 8.0 Hz, 1H), 7.18−7.25 (m, 4H), 7.28−7.32 (m, 6H), 7.44−7.48 (m, 7H), 7.60 (d, J = 4.0 Hz, 1H), 7.70 (d, J = 8.0 Hz, 1H), 7.98 (d, J = 8.0 Hz, 1H), 8.15 (s, 1H).

Methyl 2-(4-(Methoxycarbonyl)benzyloxy)-5-(tritylthiomethyl)benzoate (25d). Compound **25d** was prepared as described for the preparation of **25a** except methyl 4-(bromomethyl)benzoate was used in place of benzyl bromide: white solid (63% yield): ^{1}H NMR ($CDCl_3$) δ 3.29 (s, 2H), 3.92 (s, 3H), 3.94 (s, 3H) 5.17 (s, 2H), 6.87 (d, J = 8.5 Hz, 1H), 7.20−7.33 (m, 10 H), 7.44−7.47 (m, 7H), 7.60 (d, J = 2.3 Hz, 1H), 7.70 (dt, J = 7.7, 1.7 Hz, 1H), 7.98 (dt, J = 7.8, 1.2 Hz, 1H), 8.15 (s, 1H).

2-(Benzyloxy)-5-(tritylthiomethyl)benzoic Acid (26a). To a solution of **25a** (0.300 g, 0.57 mmol) in dioxane (10 mL) and water (10 mL) was added sodium hydroxide (0.090 g, 2.26 mmol). The mixture was heated at reflux for 3 h. The reaction was quenched with 10% $KHSO_4$, and then the mixture was evaporated to dryness. The residue was dissolved in EtOAc (50 mL), and the organic layer was washed with 10% $KHSO_4$. The organic layer was washed with water (50 mL), dried over $MgSO_4$, filtered, and concentrated. The residue was purified by a silica gel column chromatography (20% EtOAc in hexanes) to give 0.26 g of **26a** as a white solid (89% yield): ^{1}H NMR (DMSO-d_6) δ 3.26 (s, 2H) 5.16 (s, 2H) 7.07 (d, J = 8.8 Hz, 1H) 7.14−7.23 (m, 1H) 7.15−7.43 (m, 19H) 7.43−7.58 (m, 2H).

2-(2-Carboxybenzyloxy)-5-(tritylthiomethyl)benzoic Acid (26b). Compound **26b** was prepared as described for the preparation of **26a** except **25b** was used in place of **25a**: white powder (100% yield); ^{1}H NMR (DMSO-d_6) δ 3.26 (s, 2H), 5.49 (s, 2H), 6.97 (d, J = 8.6 Hz, 1H), 7.21−7.45 (m, 18H), 7.60 (dt, J = 1.4, 7.7 Hz, 1H), 7.87 (d, J = 8.0 Hz, 1H), 7.96 (dd, J = 1.1, 8.0 Hz, 1H), 12.88 (br s, 2H).

2-(3-Carboxybenzyloxy)-5-(tritylthiomethyl)benzoic Acid (26c). Compound **26c** was prepared as described for the preparation of **26a** except **25c** was used in place of **25a**: off-white solid (88% yield); ^{1}H NMR (DMSO-d_6) δ 3.26 (s, 2H), 5.24 (s, 2H), 7.07 (d, J = 8.5 Hz, 1H), 7.19 (dd, J = 2.3, 8.7 Hz, 1H), 7.24−7.30 (m, 3H), 7.34−7.39 (m, 13H), 7.51 (t, J = 7.7 Hz, 1H), 7.72 (dt, J = 7.8, 1.4 Hz, 1H), 7.87 (dt, J = 7.8, 1.4 Hz, 1H), 8.06 (s, 1H), 12.88 (br s, 2H).

2-(4-Carboxybenzyloxy)-5-(tritylthiomethyl)benzoic Acid (26d). Compound **26d** was prepared as described for the preparation of **26a** except **25d** was used in place of **25a**. Instead of column chromatography, the crude material was recrystallized from hot methanol: white powder (41% yield); ^{1}H NMR (DMSO-d_6) δ 3.26 (s, 2H), 5.25 (s, 2H), 7.06 (d, J = 8.6 Hz, 1H), 7.20 (dd, J = 2.4, 8.6 Hz, 1H), 7.27 (m, 3H), 7.37 (m, 12H), 7.40 (d, J = 2.4 Hz, 1H), 7.59 (d, J = 8.5 Hz, 2H), 7.95 (d, J = 8.5 Hz, 2H), 12.87 (brs, 2H).

2-(Benzyloxy)-5-(mercaptomethyl)benzoic Acid (27a). To a solution of **26a** (0.250 g, 0.48 mmol) in dichloromethane (2 mL) was dropwise added triisopropylsilane (0.12 mL, 0.58 mmol) followed by TFA (0.5 mL). The reaction mixture slowly turned colorless over the next 0.5 h and was allowed to continue stirring for an additional 3 h. The solvent was evaporated to dryness and the residue was purified by a silica gel column chromatography (50% EtOAc in hexanes) to give 0.094 g of **27a** as a white solid (71% yield): mp 132−138; ^{1}H NMR ($CDCl_3$) δ 1.79 (t, J = 7.8 Hz, 1H), 3.74 (d, J = 7.8 Hz, 2H), 5.30 (s, 2H), 7.10 (d, J = 8.6 Hz, 1H), 7.37−7.49 (m, 5H), 7.55 (dd, J = 8.59, 2.27 Hz, 1 H), 8.14 (d, J = 2.3 Hz, 1H). Anal. Calcd for $C_{15}H_{14}SO_3 \cdot 0.3H_2O$: C, 64.40; H, 5.26; S, 11.46. Found: C, 64.73; H, 5.25; S, 11.25.

2-(2-Carboxybenzyloxy)-5-(mercaptomethyl)benzoic Acid (27b). Compound **27b** was prepared as described for the preparation of **27a** except **26b** was used in place of **26a**. The crude material was purified by trituration with hot EtOAc to give **27b** as a white solid (75% yield): mp 196−203 °C; ^{1}H NMR (DMSO-d_6) δ 2.91 (t, J = 7.8 Hz, 1H), 3.72 (d, J = 7.8 Hz, 2H), 5.51 (s, 2H), 7.03 (d, J = 8.6 Hz, 1H), 7.46 (m, 2H), 7.61 (dt, J = 1.4, 7.6 Hz, 1H), 7.68 (d, J = 2.4 Hz, 1H), 7.90 (dd, J = 0.9, 7.8 Hz, 1H), 7.96 (dd, J = 1.3, 7.8 Hz, 1H), 12.93 (br s, 2H); ^{13}C NMR (DMSO-d_6) δ 26.8, 68.2, 113.6, 121.3, 127.2, 127.4, 128.4, 130.6, 130.7, 132.4, 132.9, 133.7, 138.8, 155.9, 167.2, 167.9. Anal. Calcd for $C_{16}H_{14}O_5S \cdot 1.7NaCl$: C, 46.01; H, 3.38; S, 7.68. Found: C, 46.06; H, 3.37; S, 7.65.

2-(3-Carboxybenzyloxy)-5-(mercaptomethyl)benzoic Acid (27c). Compound **27c** was prepared as described for the preparation of **27a** except **26c** was used in place of **26a**: white solid (17% yield); mp 215−217 °C; ^{1}H NMR (DMSO-d_6) δ 2.89 (t, J = 7.7 Hz, 1H), 3.71 (d, J = 7,6 Hz, 2H), 5.26 (s, 2H), 7.14 (d, J = 8.5 Hz, 1H), 7.44 (dd, J = 2.4, 8.6 Hz, 1H), 7.52 (t, J = 7.8 Hz, 1H), 7.64 (d, J = 2.3 Hz, 1H), 7.74 (d, J = 7.8 Hz, 1H), 7.88 (dt, J = 7.7, 1.3 Hz, 1H), 8.07 (s, 1H), 12.92 (br s, 2H); ^{13}C NMR (DMSO-d_6) δ 26.7, 69.2, 114.1, 121.8, 127.9, 128.6, 128.7, 130.5, 131.0, 131.4, 132.6, 133.8, 137.7, 155.7, 167.2 (2C). Anal. Calcd for $C_{16}H_{14}S_1O_5 \cdot 0.4H_2O$: C, 59.03; H, 4.58; S, 9.85. Found: C, 58.81; H, 4.21; S, 9.84.

2-(4-Carboxybenzyloxy)-5-(mercaptomethyl)benzoic Acid (27d). Compound **27d** was prepared as described for the preparation of **27a** except **26d** was used in place of **26a**: white solid (69% yield); mp 181−193 °C; ^{1}H NMR (DMSO-d_6) δ 2.89 (t, J = 7.7 Hz, 1H), 3.72 (d, J = 7.5 Hz, 2H), 5.27 (s, 2H), 7.12 (d, J = 8.6 Hz, 1H), 7.43 (dd, J = 2.4, 8.6 Hz, 1H), 7.60 (d, J = 8.5 Hz, 2H), 7.65 (d, J = 2.3 Hz, 1H), 7.95 (d, J = 8.4 Hz, 2H), 12.87 (br s, 2H); ^{13}C NMR (DMSO-d_6) δ 26.8, 69.2, 114.2, 122.0, 127.1, 129.6, 130.4, 130.7, 132.8, 134.0, 142.4, 155.8, 167.2, 167.3. Anal. Calcd for $C_{16}H_{14}SO_5 \cdot 0.3H_2O$: C, 59.36; H, 4.55; S, 9.90. Found: C, 59.45; H, 4.64; S, 9.66.

2,2-Dimethyl-5-vinyl-4*H*-benzo[*d*][1,3]dioxin-4-one (29). To a solution of 2,2-dimethyl-4-oxo-4*H*-benzo[*d*][1,3]dioxin-5-yl trifluoromethanesulfonate **28** (16.3 g, 50 mmol) in 1,2-dimethoxyethane (200 mL) and water (50 mL) were added tetrakis(triphenylphosphine) palladium (5.77 g, 5.00 mmol), potassium carbonate (13.9 g, 100 mmol), and vinylboronic anhydride pyridine complex (14.4 g, 60.0 mmol) under argon atmosphere. The reaction mixture was heated at 80 °C for 5 h. The reaction mixture was diluted with EtOAc (200 mL). The organic layer was separated, washed with water (75 mL × 2), dried over Na_2SO_4, filtered, and concentrated in vacuo. The crude residue was purified by silica gel column chromatography (3% EtOAc in hexanes) to afford 8.50 g of **29** as a yellow syrup (83% yield): ^{1}H NMR ($CDCl_3$) δ 1.72 (s, 6H), 5.43 (d, J = 8.8 Hz, 1H), 5.71 (d, J = 13.6 Hz, 1H), 6.89 (d, J = 6.0 Hz, 1H), 7.27 (d, J = 7.2 Hz, 1H), 7.47 (t, J = 6.4 Hz, 1H), 7.73 (dd, J = 8.8, 13.6 Hz, 1H).

***S*-2-(2,2-Dimethyl-4-oxo-4*H*-benzo[*d*][1,3]dioxin-5-yl)ethyl Ethanethioate (30).** To a solution of **29** (23.0 g, 113 mmol) and thioacetic acid (25.7 g, 338 mmol) in toluene (350 mL) was added AIBN (18.5 g, 113 mmol) at 80 °C. After the mixture was stirred for 4 h, saturated aqueous $NaHCO_3$ (100 mL) and EtOAc (300 mL) were poured into the reaction mixture. The organic layer was washed with saturated aqueous $NaHCO_3$ (100 mL × 3), water, and brine, dried over Na_2SO_4, and concentrated in vacuo. The crude material was purified by silica gel column chromatography (1% EtOAc in hexanes) to afford 17.5 g of **30** as a low melting solid (55% yield): ^{1}H NMR ($CDCl_3$) δ 1.71 (s, 6H), 2.30 (s, 3H), 3.18 (t, J = 7.2 Hz, 2H), 3.34 (t, J = 7.2 Hz, 2H), 6.86 (d, J = 8.4 Hz, 1H), 6.99 (d, J = 7.6 Hz, 1H), 7.43 (t, J = 8.0 Hz, 1H).

2-Hydroxy-6-(2-mercaptoethyl)benzoic Acid (31). To a solution of **30** (10.0 g, 35.6 mmol) in a mixture of methanol and

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water (200 mL, 1:1) were added sodium hydroxide (5.71 g, 143 mmol) and one spatula amount of tris(2-carboxyethyl)phosphine (TCEP). The mixture was heated at 65 °C for 3.5 h. The solvents were removed in vacuo. The resulting residue was dissolved in EtOAc and acidified with aqueous 10% $KHSO_4$. The organic phase was washed with brine, dried over Na_2SO_4, and concentrated to give 6.55 g of **31** as a brown oil (93% crude yield). The material was used in subsequent reaction as is without purification: ^{1}H NMR ($CDCl_3$) δ 1.45 (t, J = 8.0 Hz, 1H), 2.83 (t, J = 7.8 Hz, 2H), 3.30 (t, J = 7.8 Hz, 2H), 6.81 (dd, J = 1.0, 7.4 Hz, 1H), 6.93 (dd, J = 1.1, 8.4 Hz, 1H), 7.42 (dd, J = 7.6, 8.4 Hz, 1H), 10.49 (bs, 1H).

8-Hydroxyisothiochroman-1-one (32). A solution of **31** (6.51 g, 32.8 mmol) in toluene (200 mL) was heated at 110 °C for 24 h in the presence of *p*-TsOH (1.70 g, 9.85 mmol). The reaction mixture was concentrated, and the residue was taken up by EtOAc and saturated aqueous $NaHCO_3$. The organic layer was subsequently washed with 10% $KHSO_4$ and brine, dried over Na_2SO_4, and concentrated in vacuo. The resulting brown oil was purified by silica gel column chromatography (20% EtOAc in hexanes) to provide 4.23 g of **32** as a brown semisolid (71% yield): ^{1}H NMR ($CDCl_3$) δ 3.23 (m, 2H), 3.27 (m, 2H), 6.69 (m, 1H), 6.88 (m, 1H), 7.33 (dd, J = 7.6, 8.6 Hz, 1H), 11.59 (s, 1H).

1-Oxoisothiochroman-8-yl Trifluoromethanesulfonate (33). To a solution of **32** (3.02 g, 16.8 mmol) in dichloromethane (30 mL) was added pyridine (3.00 mL, 36.9 mmol), followed by the addition of 1.0 M solution of trifluoromethanesulfonic anhydride in dichloromethane (18.4 mL, 18.4 mmol) at 0 °C. The reaction mixture was stirred at 0 °C for 2.5 h, and an additional 0.3 equiv of 1.0 M of triflic anhydride and 0.3 equiv of pyridine were added. After 1.5 h, more triflic anhydride (1 mL) was added and stirring continued for 30 min. The reaction was quenched with 1 N HCl. The organic layer was washed with brine, dried over Na_2SO_4, and concentrated in vacuo. The crude material was recrystallized from EtOAc/hexanes to give 3.08 g of **33** as the first crop. The mother liquor was purified by silica gel column chromatography (20−50% EtOAc in hexanes) to give 0.41 g of **33** as the second crop: light brown solid (3.49 g, 67% combined yield); ^{1}H NMR (CD_3OD) δ 3.29 (s, 4H), 7.27 (d, J = 8.1 Hz, 1H), 7.34 (d, J = 7.1 Hz, 1H), 7.56 (t, J = 7.9, 1H).

8-Phenylisothiochroman-1-one (34a). To a solution of **33** (0.500 g, 1.60 mmol) in toluene (10 mL) were added cesium carbonate (1.04 g, 3.19 mmol), phenylboronic acid **15a** (0.235 g, 1.92 mmol), and tetrakis(triphenylphosphine)palladium (0.092 g, 0.08 mmol). The mixture was heated at 110 °C for 2.5 h. After cooling, the mixture was filtered through a pad of Celite and concentrated in vacuo. Trituration of the residual solid with a mixture of EtOAc and hexanes gave 0.220 g of **34a** as a yellow solid (57% yield): ^{1}H NMR ($CDCl_3$) δ 3.28 (m, 4H), 7.25−7.30 (m, 4H), 7.34−7.41 (m, 3H), 7.47 (t, J = 7.6 Hz, 1H).

Methyl 2-(1-Oxoisothiochroman-8-yl)benzoate (34b). Compound **34b** was prepared as described for the preparation of **34a** except 2-(methoxycarbonyl)phenylboronic acid **15b** was used in place of **15a**. The crude material was purified by Biotage Isolera One using EtOAc/hexanes as eluent: viscous oil (51% yield); ^{1}H NMR ($CDCl_3$) δ 3.27 (m, 4H), 3.68 (s, 3H), 7.13 (dd, J = 1.3, 7.6 Hz, 1H), 7.18 (m, 1H), 7.26 (m, 1H), 7.41 (m, 1H), 7.47 (t, J = 7.6 Hz, 1H), 7.53 (m, 1H), 8.00 (m, 1H).

Ethyl 3-(1-Oxoisothiochroman-8-yl)benzoate (34c). Compound **34c** was prepared as described for the preparation of **34a** except 3-(ethoxycarbonyl)phenylboronic acid was used in place of **15a**. The crude material was purified by Biotage Isolera One using EtOAc/hexanes as eluent: yellow oil (88% yield); ^{1}H NMR ($CDCl_3$) δ 1.39 (t, J = 7.1 Hz, 3H), 3.29 (m, 4H), 4.37 (q, J = 7.1 Hz, 2H), 7.28−7.30 (m, 2H), 7.44−7.46 (m, 2H), 7.49 (t, J = 7.6 Hz, 1H), 7.97 (m, 1H), 8.02 (m, 1H).

Methyl 4-(1-Oxoisothiochroman-8-yl)benzoate (34d). Compound **34d** was prepared as described for the preparation of **34a** except 4-(methoxycarbonyl)phenylboronic acid **15d** was used in place of **15a**. The compound was purified by recrystallization from EtOAc/hexanes: off-white solid (49% yield); ^{1}H NMR ($CDCl_3$) δ 3.30 (m, 4H), 3.93 (s, 3H), 7.28−7.31 (m, 2H), 7.34 (m, 1H), 7.36 (m, 1H), 7.49 (t, J = 7.6 Hz, 1H), 8.05 (m, 1H), 8.07 (m, 1H).

3-(2-Mercaptoethyl)biphenyl-2-carboxylic Acid (35a). To a solution of **34a** (0.111 g, 0.47 mmol) in a mixture of 1,4-dioxane and water (5 mL, 1:1) was added 1 M NaOH (1 mL), and the resulting mixture was stirred at 100 °C for 2 h. The mixture was subsequently treated with a spatula of TCEP until complete disappearance of the corresponding disulfide byproduct. The solvents were removed, and the residual material was taken up in aqueous 10% $KHSO_4$ and EtOAc. The organic layer was washed with brine, dried over Na_2SO_4, and concentrated in vacuo. The resulting residue was triturated with 10% EtOAc in hexanes to give 0.068 g of **35a** as a white solid (56% yield): mp 153−155 °C; ^{1}H NMR ($CDCl_3$) δ 1.47 (t, J = 8.1, 1H), 2.86 (m, 2H), 3.06 (m, 2H), 7.29 (m, 1H), 7.31 (m, 1H), 7.39−7.42 (m, 5H), 7.46 (t, J = 7.6 Hz, 1H). Anal. Calcd for $C_{15}H_{14}O_2S$: C, 69.74; H, 5.46; S, 12.41. Found: C, 69.87; H, 5.47; S, 12.23.

3-(2-Mercaptoethyl)biphenyl-2,2′-dicarboxylic Acid (35b). Compound **35b** was prepared as described for the preparation of **35a** except **34b** was used in place of **34a**. The crude material was purified by reverse phase preparative HPLC: white solid (61% yield); mp 185−187 °C; ^{1}H NMR ($CDCl_3$) δ 1.43 (t, J = 8.1, 1H), 2.83 (m, 2H), 3.04 (m, 2H), 7.04 (dd, J = 1.3, 7.6 Hz, 1H), 7.28 (m, 1H), 7.40 (t, J = 7.7 Hz, 1H), 7.47 (m, 1H), 7.56 (m, 1H), 7.91 (dd, J = 1.3, 7.6 Hz, 1H). Anal. Calcd for $C_{16}H_{14}O_4S{\cdot}0.2H_2O$: C, 62.81; H, 4.74; S, 10.48. Found: C, 62.60; H, 4.61; S, 10.53.

3-(2-Mercaptoethyl)biphenyl-2,3′-dicarboxylic Acid (35c). Compound **35c** was prepared as described for the preparation of **35a** except **34c** was used in place of **34a**. The crude material was purified by recrystallization from EtOAc/hexanes: white powder (68% yield); mp 165−167 °C; ^{1}H NMR ($CDCl_3$) δ 1.52 (t, J = 8.1 Hz, 1H), 2.90 (m, 2H), 3.11 (m, 2H), 7.38 (m, 2H), 7.49−7.58 (m, 2H), 7.70 (m, 1H), 7.97 (m, 1H), 8.30 (s, 1H). Anal. Calcd for $C_{16}H_{14}O_4S{\cdot}0.25AcOH$: C, 62.45; H, 4.76; S, 10.10. Found: C, 62.33; H, 4.85; S, 10.17.

3-(2-Mercaptoethyl)biphenyl-2,4′-dicarboxylic Acid (35d). Compound **35d** was prepared as described for the preparation of **35a** except **34d** was used in place of **34a**. The crude material was purified by reverse phase preparative HPLC: white powder (53% yield); mp 245−246 °C; ^{1}H NMR (DMSO-d_6) δ 2.45 (t, J = 7.8, 1H), 2.73 (m, 2H), 2.93 (m, 2H), 7.29 (dd, J = 1.3, 7.6 Hz, 1H), 7.40 (dd, J = 1.3, 7.6 Hz, 1H), 7.46 (d, J = 7.6 Hz, 1H), 7.49 (m, 1H), 7.52 (m, 1H), 7.97 (m, 1H), 7.99 (m, 1H), 13.04 (bs, 1H), 13.19 (bs, 1H). Anal. Calcd for $C_{16}H_{14}O_4S$: C, 63.56; H, 4.67; S, 10.61. Found: C, 63.28; H, 4.66; S, 10.38.

8-(Benzyloxy)isothiochroman-1-one (36a). To a solution of **32** (0.220 g, 1.22 mmol) in acetone (10 mL) were added benzyl bromide (0.310 g, 1.83 mmol) and potassium carbonate (0.250 g, 1.83 mmol). The mixture was heated at 55 °C overnight. Solids were removed by filtration, and the filtrate was concentrated in vacuo. The resulting residue was purified by silica gel column chromatography (50% EtOAc in hexanes, 1% acetic acid) to provide 0.100 g of **36a** as an off-white solid (31% yield): ^{1}H NMR ($CDCl_3$) δ 3.19 (s, 4H), 5.20 (s, 2H), 6.84 (d, J = 6.6 Hz, 1H), 6.99 (d, J = 8.6 Hz, 1H), 7.28−7.33 (m, 1H), 7.33−7.42 (m, 3H), 7.53 (d, J = 7.1 Hz, 2H).

Methyl 2-((1-Oxoisothiochroman-8-yloxy)methyl)benzoate (36b). Compound **36b** was prepared as described for the preparation of **36a** except methyl 2-(bromomethyl)benzoate was used in place of benzyl bromide. The resulting residue was recrystallized from EtOAc/hexanes: yellow crystals (40% yield); ^{1}H NMR ($CDCl_3$) δ 3.22 (s, 4H), 3.93 (s, 3H), 5.59 (s, 2H), 6.86 (d, J = 6.8 Hz, 1H), 7.12 (d, J = 8.3 Hz, 1H), 7.34−7.46 (m, 2H), 7.67 (t, J = 7.0 Hz, 1H), 8.05 (d, J = 6.8 Hz, 1H), 8.23 (d, J = 7.8 Hz, 1H).

Methyl 3-((1-Oxoisothiochroman-8-yloxy)methyl)benzoate (36c). Compound **36c** was prepared as described for the preparation of **36a** except methyl 3-(bromomethyl)benzoate was used in place of benzyl bromide: clear oil (61% yield); ^{1}H NMR ($CDCl_3$) δ 3.20 (s, 3H), 3.93 (s, 4 H), 5.22 (s, 2H), 6.86 (d, J = 7.6 Hz, 1H), 6.97 (d, J = 8.6 Hz, 1H), 7.33−7.41 (m, 1H), 7.49 (t, J = 7.7 Hz, 1H), 7.87 (d, J = 7.6 Hz, 1H), 7.98 (d, J = 7.8 Hz, 1H), 8.10 (s, 1H).

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Methyl 4-((1-Oxoisothiochroman-8-yloxy)methyl)benzoate (36d). Compound **36d** was prepared as described for the preparation of **36a** except methyl 4-(bromomethyl)benzoate was used in place of benzyl bromide. The crude material was recrystallized from EtOAc/hexanes: beige powder (39% yield); ^{1}H NMR ($CDCl_3$) δ 3.21 (s, 3H), 3.92 (s, 4H), 5.24 (s, 2H), 6.87 (d, J = 7.1 Hz, 1H), 6.96 (d, J = 8.3 Hz, 1H), 7.38 (t, J = 8.0 Hz, 1H), 7.62 (d, J = 8.6 Hz, 2H), 8.07 (d, J = 8.3 Hz, 2H).

2-(Benzyloxy)-6-(2-mercaptoethyl)benzoic Acid (37a). To a solution of **36a** (0.090 g, 0.34 mmol) in H_2O/dioxane (1.0 mL, 1:1) was added sodium hydroxide (0.040 g, 1.01 mmol). The mixture was heated at 100 °C for 2.5 h. A spatula of TCEP was added, and the mixture was stirred for 15 min at room temperature. Solvents were removed in vacuo, and the residual material was taken up in aqueous 10% $KHSO_4$ and EtOAc. The organic layer was washed with brine, dried over Na_2SO_4, and concentrated. The resulting residue was purified by preparative HPLC to give 0.024 g of **37a** as white crystals (25% yield): mp 54−56 °C; ^{1}H NMR ($CDCl_3$) δ 1.45 (t, J = 8.2 Hz, 1H), 2.82 (q, J = 7.6, 15.2 Hz, 2H), 3.11 (t, J = 7.8 Hz, 2H), 5.19 (s, 2H), 6.95 (d, J = 8.6 Hz, 2H), 7.33−7.43 (m, 6H). Anal. Calcd for $C_{16}H_{16}O_3S·0.2H_2O$: C, 65.82; H, 5.66; S, 10.98. Found: C, 65.94; H, 5.57; S, 10.70.

2-(2-Carboxybenzyloxy)-6-(2-mercaptoethyl)benzoic Acid (37b). Compound **37b** was prepared as described for the preparation of **37a** except **36b** was used in place of **36a**: white powder (13% yield); mp 171−173 °C; ^{1}H NMR (CD_3OD) δ 2.75 (m, 2H), 2.93 (m, 2H), 5.55 (s, 2H), 6.93 (dd, J = 2.5, 7.6 Hz, 2H), 7.30 (t, J = 8.3 Hz, 1H), 7.40 (t, J = 7.8 Hz, 1H), 7.57 (dt, J = 1.3, 7.8 Hz, 1H), 7.82 (d, J = 7.8 Hz, 1H), 8.06 (dd, J = 1.0, 7.8 Hz, 1H). Anal. Calcd for $C_{17}H_{16}O_5S·0.2H_2O$: C, 60.77; H, 4.92; S, 9.54. Found: C, 60.86; H, 4.85; S, 9.32.

2-(3-Carboxybenzyloxy)-6-(2-mercaptoethyl)benzoic Acid (37c). Compound **37c** was prepared as described for the preparation of **37a** except **36c** was used in place of **36a**. The crude material was recrystallized from EtOAc/hexanes: yellow powder (61% yield); mp 142−147 °C; ^{1}H NMR (CD_3OD) δ 2.74 (t, J = 7.6 Hz, 2H), 2.90 (dd, J = 5.6, 8.3 Hz, 2H), 5.19 (s, 2H), 6.93 (d, J = 7.6 Hz, 1H), 7.01 (d, J = 8.1 Hz, 1H), 7.31 (t, J = 7.8 Hz, 1H), 7.48 (t, J = 7.7 Hz, 1H), 7.71 (d, J = 8.1 Hz, 1H), 7.97 (d, J = 7.8 Hz, 1H), 8.12 (s, 1H). Anal. Calcd for $C_{17}H_{16}O_5S$: C, 61.43; H, 4.85; S, 9.65. Found: C, 61.31; H, 4.86; S, 9.36.

2-(4-Carboxybenzyloxy)-6-(2-mercaptoethyl)benzoic Acid (37d). Compound **37d** was prepared as described for the preparation of **37a** except **36d** was used in place of **36a**. The crude material was recrystallized from EtOAc/dichloromethane: off-white powder (13% yield); mp 193−196 °C; ^{1}H NMR (CD_3OD) δ 2.75 (t, J = 8.2 Hz, 2H), 2.92 (t, J = 7.7 Hz, 2H), 5.23 (s, 2H), 6.93 (d, J = 7.6 Hz, 1H), 6.98 (d, J = 8.1 Hz, 1H), 7.32 (t, J = 8.0 Hz, 1H), 7.57 (d, J = 7.6 Hz, 2H), 8.03 (d, J = 8.1 Hz, 2H). Anal. Calcd for $C_{17}H_{16}O_5S·0.4H_2O$: C, 60.13; H, 4.99; S, 9.44. Found: C, 60.03; H, 4.80; S, 9.16.

Pharmacological Studies. The GCPII assay was carried out as outlined previously.[21] The Ames test was performed by Covance Laboratories, Vienna, VA. The hERG assay was conducted by Zenas Technologies, New Orleans, LA. Receptor binding assays were performed by MDS Pharma Services, Bothell, WA. Details of the pharmacokinetics study will be reported elsewhere. The chronic constrictive injury models were performed following the procedure reported by Bennett's group[23] and will be detailed elsewhere.

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Notes

The authors declare no competing financial interest.

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ABBREVIATIONS USED

GCPII, glutamate carboxypeptidase II; NAAG, *N*-acetylaspartylglutamate; 2-MPPA, 2-(3-mercaptopropyl)pentanedioic acid; ALS, amyotrophic lateral sclerosis; CMBA, 3-(2-carboxy-5-mercaptopentyl)benzoic acid; AUC, area under the curve

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