Ester and Amide Prodrugs of Ibuprofen and Naproxen: Synthesis, Anti-inflammatory Activity, and Gastrointestinal Toxicity

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Abstract □ Ester and amide prodrugs of ibuprofen (**1**) and naproxen (**16**) were synthesized and evaluated for anti-inflammatory activity and gastrointestinal toxicity. The chemical structure of the prodrugs was varied in terms of lipophilicity and reactivity toward hydrolysis. Inhibition of acetic acid-induced writhing in mice indicated that prodrugs **7**, **15**, **19**, and **20** exhibited significantly better activity ($p < 0.01$) than the parent compounds. The average number of ulcers formed in the gastric mucosa following oral administration of **1** and **16** and prodrugs **5**, **18**, **21**, and **22** was determined in rats. All prodrugs, except the glycine amide **21**, were significantly less irritating to the gastric mucosa than either **1** or **16**.

Gastrointestinal side effects constitute the most frequent of all the adverse reactions of nonsteroidal anti-inflammatory drugs (NSAIDs).[1] These reactions range in both severity and frequency from relatively mild to the more serious and potentially life-threatening states, such as gastrointestinal ulceration and hemorrhage.[2,3] The major factor in the development of gastrointestinal ulceration and hemorrhage induced by NSAIDs is the inhibition of prostaglandin synthesis. Endogenous prostaglandins are known to have a cytoprotective action on the gastric mucosa. Prostaglandins help regulate acid secretion and maintain mucosal integrity against stress, a variety of chemicals, and thermal injury.[4,5]

Most workers generally accept the fact that gastrointestinal lesions produced by NSAIDs are the result of two different mechanisms: (1) a direct contact effect, and (2) a generalized systemic effect which may be manifested after iv dosing.[6] The direct contact effect can be attributed to a combination of local irritation produced by the acidic group of the NSAID and local inhibition of prostaglandin synthesis in the gastrointestinal tract.

In a study by Cioli et al.,[7] the importance of the direct contact effect in the gastrointestinal toxicity of ibuprofen (**1**) was examined in rats. The results of this study showed that **1** exerted a greater toxicity on the stomach and the intestine by the oral route than by the iv route. The two routes of administration showed the same anti-inflammatory activity on carrageenan-induced edema in the hind paw of the rat. The results of this study suggest that direct tissue contact of the NSAID plays an important role in the production of gastrointestinal lesions.

The use of prodrugs to temporarily mask the acidic group of NSAIDs has been postulated as an approach to decrease the gastrointestinal toxicity due to the direct contact effect.[8–10] Ester or amide prodrugs of NSAIDs should exhibit decreased toxicity since they neither possess a free carboxylic acid group, nor do they inhibit prostaglandin biosynthesis.

Considerable research has been directed at designing prodrugs of NSAIDs, with the aim of reducing their gastrointestinal toxicity.[8–19] The toxicological and pharmacological profile of ibuprofen guaiacol ester has been investigated.[11] The ester exhibited less toxicity than **1** on both the gastric and intestinal mucosa. Reduced gastrointestinal effects do not appear to be due to decreased systemic activity of the ester. The guaiacol ester was as active as **1** on both edema and fever at equimolar doses. However, one drawback of the guaiacol ester is that, after oral administration, the peak concentration of **1** in the blood is delayed. The capacity of the esters to exhibit the full therapeutic effectiveness inherent in the parent NSAID depends on their ability to release the NSAID after absorption through the gastric mucosa. Hence, simple esters and some amides are not sufficiently labile in vivo to ensure a high rate and extent of prodrug conversion.

During the course of this investigation, Persico et al.[12] reported the synthesis and biological evaluation of several amino acid amides of tolmetin. Tolmetin glycine amide produced lower peak plasma tolmetin levels than an equivalent dose of tolmetin sodium, but plasma concentrations were sustained for a longer period of time. Thus, the glycine amide represents a potential approach for sustained release of NSAIDs.

Although considerable research has been directed at designing prodrugs of NSAIDs with reduced gastrointestinal toxicity, none of these approaches has resulted in an ideal prodrug. Bundgaard and Nielsen[8] have stated that, in order for prodrugs of NSAIDs to exhibit wide utility, they should satisfy a number of criteria: (1) the prodrugs should be readily hydrolyzed following absorption to release the parent drug; (2) the prodrugs should have adequate water solubility and lipophilicity to assure absorption by the oral route; and (3) the prodrugs should be stable toward gastrointestinal enzymes prior to absorption in the gastrointestinal tract. Pharmacological data to establish anti-inflammatory activity of the prodrugs are unavailable in most of these cases. Additionally, several of these prodrugs show poor aqueous solubility. Intravenous formulations of NSAIDs have been demonstrated to be significantly less ulcerogenic than oral formulations.[7,20] Most of the prodrugs of NSAIDs reported in literature have poor or limited water solubility. Good aqueous solubility is essential for the development of iv dosage forms of prodrugs of NSAIDs. Furthermore, adequate aqueous solubility is required for acceptable bioavailability.

The purpose of this investigation was to synthesize and to evaluate the anti-inflammatory activity and gastrointestinal toxicity of ibuprofen (**1**) and naproxen (**16**) prodrugs. The chemical structure of the prodrugs was varied in terms of lipophilicity and reactivity toward hydrolysis. The stability of the individual prodrugs in human serum and aqueous media and the bioavailability of **5** compared with **1** will be the subject of a separate report.

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Experimental Section

Melting points were determined on a Thomas-Hoover melting point apparatus and are uncorrected. The IR spectra were recorded on a Nicolet 5MX FT spectrometer, and NMR spectra were recorded as 6% weight/volume solutions on a JEOL FX-90Q spectrometer. Chemical shifts are reported in parts per million (δ) relative to tetramethylsilane (1%) as the internal standard. The HPLC studies were performed on an Altex 100 pump attached to an ISCO model V[40] variable wavelength detector and a Rheodyne injector with a 20-μL loop injection valve. In some cases, an HPLC system consisting of a Varian 5000 liquid chromatograph connected to a Varian UV-50 variable wavelength detector and a Waters 740 data module was used. A reversed-phase LiChrosorb RP-8 column was used for the analysis of all compounds. Chromatograms of all prodrugs showed no detectable amounts of parent drug. Optical rotations were measured on a Perkin-Elmer 241 polarimeter. Analytical data were obtained from Desert Analytics, Inc., Tucson, AZ and Oneida Research Services, Inc., Whitesboro, NY. Ibuprofen (1; lot # 12023) was obtained from Boots Pharmaceuticals, Inc., Shreveport, LA, as a gift and was recrystallized from ethanol:water prior to use. Naproxen [16; $[\alpha]_D^{25°} = +66°$ (c 1, $CHCl_3$)] was obtained from Aldrich Chemical Company and was used as received. Tissue examinations were performed with a Bausch and Lomb (2 × 2) binocular magnifier.

Synthesis of Aminophenyl Esters—The synthesis of 4-aminophenyl 2-(4-isobutylphenyl)propionate hydrochloride (2) is representative of the general procedure. Ibuprofen (1; 5.0 g, 24.0 mmol) was dissolved in CH_3CN (150 mL), and *p*-nitrophenol (3.36 g, 24.0 mmol) and dicyclohexylcarbodiimide (4.95 g, 24.0 mmol) were added to the solution. The reaction mixture was stirred overnight at room temperature in the presence of a drying tube. The precipitated dicyclohexylurea was filtered, and the solvent was evaporated under reduced pressure to afford a yellow oil. The oil was dissolved in ethanol (100 mL), and 0.5 g of 5% Pd/C was added. The mixture was shaken on a Parr hydrogenator for 24 h at an initial pressure of 50 psi. The catalyst was filtered, and the solvent was evaporated under reduced pressure to yield an oil. Flash chromatography using 100 g of silica gel gave 5.0 g of the free base as a yellow oil. The oil was dissolved in ethanol, and hydrogen chloride gas was passed into the solution. Removal of the precipitated solid by filtration, followed by recrystallization, yielded 3.1 g of yellow crystalline 2; IR (KBr): 1760 (C=O, ester) cm^{-1}; NMR ($CDCl_3$): δ 0.88 (d, 6 H, J = 6 Hz, $(CH_3)_2$), 1.47 (d, 3 H, J = 6 Hz, CH_3CH), 1.78 (m, 1 H, $(CH_3)_2CH$), 2.48 (d, 2 H, J = 6 Hz, CH_2Ar), 3.62 (q, 1 H, J = 6 Hz, CH_3CH), 7.17 (m, 4 H, ArH). Compounds 3 and 17 were also obtained by this method.

Synthesis of Aminoalkyl Esters and Amides—The synthesis of 2-(*N*,*N*-dimethylamino)ethyl 2-(4-isobutylphenyl)propionate hydrochloride (5) is representative of the general method.[21] A solution of 1 (5.0 g, 24.0 mmol) in dry toluene (150 mL) was heated to reflux and treated in a dropwise manner with $SOCl_2$ (16.5 mL). After refluxing for 2 h, the excess $SOCl_2$ was azeotropically removed with dry toluene under reduced pressure. The crude acid chloride was dissolved in THF (150 mL) and treated with *N*,*N*-dimethylethanolamine (4.32 g, 48.4 mmol) in a dropwise fashion. A white precipitate formed almost immediately, and stirring was continued overnight at room temperature. The precipitated *N*,*N*-dimethylethanolamine hydrochloride was removed by filtration, and the filtrate was evaporated under reduced pressure to give a yellow oil. Vacuum distillation of the free base, followed by preparation and recrystallization of the hydrochloride, gave analytically pure 5; IR (KBr): 1750 (C=O, ester) cm^{-1}; NMR ($CDCl_3$): δ 0.88 (d, 6 H, J = 6 Hz, $(CH_3)_2$), 1.52 (d, 3 H, CH_3CH), 1.83 (m, 1 H, $(CH_3)_2CH$), 2.46 (d, 2 H, J = 6 Hz, CH_2Ar), 2.59 (s, 6 H, $N(CH_3)_2$), 3.24 (t, 2 H, CH_2N), 3.75 (q, 1 H, CH_3CH), 4.56 (t, 2 H, J = 6 Hz, OCH_2), 7.15 (m, 4 H, ArH). This method was used to prepare 5–13, 18–20, and 23. In the case of 23, an analytical sample was obtained by a second distillation under reduced pressure.

2-Aminoethyl 2-(4-Isobutylphenyl)propionate Hydrochloride (4)—This compound was prepared using a mixed anhydride method.[22,23] A solution of 1 (1.92 g, 9.3 mmol) in THF (25 mL) was cooled to 0–5 °C and treated with Et_3N (0.94 g, 9.3 mmol), followed by the addition of 4-dimethylaminopyridine (DMAP; 0.11 g, 0.9 mmol). Isopropenyl chloroformate (1.12 g, 9.0 mmol) was added in a dropwise manner to the reaction mixture, and stirring was continued for 15 min, whereupon a white solid formed. The cooled solution was treated with 2-[*N*-(*tert*-butyloxycarbonyl)]aminoethanol[24] [bp 107–110 °C (1.1 mmHg)], and stirring was continued for 3 h, during which time the mixture warmed to room temperature. The precipitate was filtered, and the solvent was evaporated. The resultant oil was dissolved in $CHCl_3$ and washed successively with 1 M citric acid (3 × 50 mL), H_2O (1 × 50 mL), cold 5% NaOH (3 × 50 mL), and H_2O (1 × 50 mL). The $CHCl_3$ layer was dried (Na_2SO_4), filtered, and evaporated under reduced pressure to yield an oil. The oil was dissolved in CH_2Cl_2, and HCl gas was bubbled into the cold solution. The solvent was evaporated, and the resultant oil was triturated with Et_2O to give a white solid. Recrystallization gave pure 4; IR (KBr): 1750 (C=O, ester) cm^{-1}; NMR ($CDCl_3$): δ 0.89 (d, 6 H, J = 6 Hz, $(CH_3)_2$), 1.50 (d, 3 H, J = 6 Hz, CH_3CH), 1.77 (m, 1 H, $(CH_3)_2CH$), 2.39 (d, 2 H, J = 6 Hz, CH_2Ar), 3.26 (m, 2 H, CH_2N), 3.62 (q, 1 H, J = 6 Hz, CH_3CH), 4.09 (t, 2 H, OCH_2), 7.19 (m, 4 H, ArH).

2-(*N*-Methylamino)ethyl 2-(4-Isobutylphenyl)propionate Hydrochloride (14)—A mixture of 13 (5.0 g, 12.8 mmol) and 1 g of 5% Pd/C in 95% ethanol (200 mL) was shaken on a Parr hydrogenator at 50 psi for 24 h. The catalyst was filtered, and the solvent was evaporated under reduced pressure to afford a white solid. Recrystallization yielded 2.11 g of a white crystalline solid; IR (KBr): 1750 (C=O, ester) cm^{-1}; NMR ($CDCl_3$): δ 0.88 (d, 6 H, J = 6 Hz, $(CH_3)_2$), 1.50 (d, 3 H, J = 6 Hz, CH_3CH), 1.78 (m, 1 H, $(CH_3)_2CH$), 2.42 (d, 2 H, J = 6 Hz, CH_2Ar), 2.57 (s, 3 H, NCH_3), 3.19 (m, 2 H, CH_2N), 3.86 (q, 1 H, J = 6 Hz, CH_3CH), 4.4 (m, 2 H, OCH_2), 7.17 (m, 4 H, ArH).

2-(1-Piperazinyl)ethyl 2-(4-Isobutylphenyl)propionate Dihydrochloride (15)—A mixture of 1-(2-hydroxyethyl)piperazine (15.0 g, 115 mmol) and $NaHCO_3$ (9.67 g, 115 mmol) in DMF (100 mL) was treated in a dropwise fashion with benzyl bromide (19.7 g, 115 mmol). The reaction mixture was allowed to come to room temperature and was stirred overnight. Removal of the DMF under reduced pressure gave a residue, which was treated with H_2O (100 mL). The clear solution was extracted with CH_2Cl_2 (3 × 50 mL), and the combined CH_2Cl_2 extracts were washed with H_2O (1 × 30 mL), dried (Na_2SO_4), filtered, and evaporated to yield an oil. Vacuum distillation gave 11.5 g (45%) of 4-benzyl-1-(2-hydroxyethyl)piperazine [bp 129–134 °C (0.4 mmHg), lit[25] bp 142–143 °C (2 mmHg)].

Ibuprofen (1; 5.0 g, 24.2 mmol) was converted to its acid chloride as described for the synthesis of 5. Reaction of the acid chloride with 4-benzyl-1-(2-hydroxyethyl)piperazine (10.6 g, 48.0 mmol) yielded 7.0 g of the crude ester. Flash chromatography on 100 g of silica gel using a hexane:EtOAc (5:5) solvent system afforded 5.8 g (59%) of an oil. An ethanolic solution of the oil was saturated with HCl gas and treated with 1.2 g of 5% Pd/C. The mixture was shaken on a Parr hydrogenator at 50 °C at an initial pressure of 50 psi for 1.5 h. The catalyst was filtered, and the solvent was evaporated under reduced pressure to yield a solid. Recrystallization gave 4.8 g of 15; IR (KBr): 1750 (C=O, ester) cm^{-1}; NMR ($CDCl_3$): δ 0.89 (d, 6 H, J = 6 Hz, $(CH_3)_2CH$), 1.47 (d, 3 H, J = 6 Hz, CH_3CH), 1.77 (m, 1 H, $(CH_3)_2CH$), 2.36–2.63 (m, 12 H, CH_2Ar, CH_2N, and piperazine ring), 3.66 (q, 1 H, J = 6 Hz, CH_3CH), 4.15 (t, 2 H, OCH_2), 7.15 (m, 4 H, ArH).

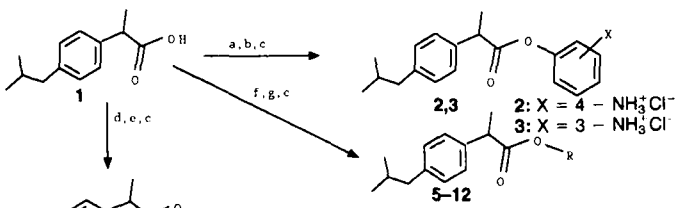
(+)*N*-[2-(6-Methoxy-2-naphthalyl)propionyl]glycine (21)—The synthesis of this compound was accomplished using a procedure reported by Sharma et al.[26] A solution of 16 (2.0 g, 9 mmol) and *N*-hydroxysuccinimide (1.1 g, 10 mmol) in THF (150 mL) was cooled to 0–5 °C under a N_2 atmosphere and treated with dicyclohexylcarbodiimide (2.3 g, 10 mmol). The reaction mixture was stirred at 0–5 °C for 3 h and allowed to warm to room temperature overnight. The precipitated dicyclohexylurea was filtered, and the THF solution of the succinimido ester was treated with glycine benzyl ester (1.5 g, 9 mmol) and DMAP (0.14 g, 0.9 mmol). After stirring overnight, the solvent was removed under reduced pressure to yield a solid residue. The residue was dissolved in CH_2Cl_2 (100 mL) and washed successively with 1 M HCl (1 × 50 mL), 4% NaOH (1 × 50 mL), and H_2O (1 × 50 mL). The organic phase was separated, dried (Na_2SO_4), filtered, and evaporated to yield a white solid. The solid was dissolved in EtOH (120 mL) and shaken for 36 h on a Parr hydrogenator at 50 psi, using 0.5 g of 5% Pd/C as the catalyst. The catalyst was filtered, and the solvent was evaporated to yield an oil. The oil was dissolved in 1 M NaOH (100 mL) and extracted with CH_2Cl_2 (2 × 50 mL). The aqueous phase was acidified with 6 M HCl to give a white solid. Recrystallization afforded 1.0 g of a white crystalline solid; IR (KBr): 1740 (C=O, acid), 1650 (C=O, amide) cm^{-1}; NMR (DMSO-d_6): δ 1.45 (d, 3 H, J = 6 Hz, CH_3CH), 3.52 (q, 1 H, CH_3CH), 3.75 (d, 2 H, $NHCH_2$), 3.85 (s, 3 H, OCH_3), 7.50 (m, 6 H, ArH), 8.25 (t, 1 H, NH).

***N*-[2-(4-Isobutylphenyl)propionyl]glycine (22)**—Glycine benzyl ester hydrochloride (3.96 g, 24.0 mmol) was suspended in THF (150 mL) and treated with Et_3N (2.42 g, 24.0 mmol). After stirring for 2

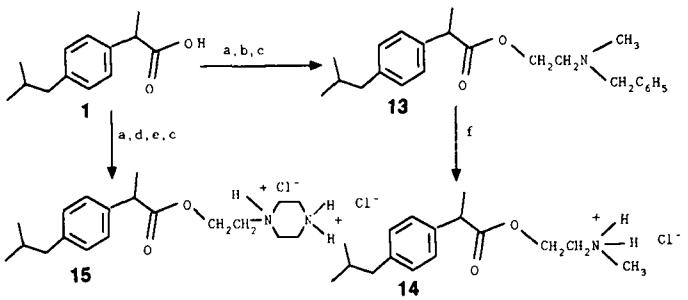
Table I—Ulcerogenic Activity in Rats[a]

Compound[b]	Ulcers[c]
1	12.57 ± 11.88
5	1.50 ± 0.84[d]
16	4.00 ± 2.89
18	1.66 ± 0.52[d]
21	2.33 ± 2.16
22	2.00 ± 1.41[d]

[a] n = 6. [b] Ibuprofen (**1**; 150 mg, 0.727 mmol/kg/day); naproxen (**16**; 40 mg, 0.174 mmol/kg/day); prodrugs **5** and **22** (0.727 mmol/kg/day); prodrugs **18** and **21** (0.174 mmol/kg/day). [c] Average number of ulcers >0.5 mm formed ± SD. [d] $p < 0.01$.



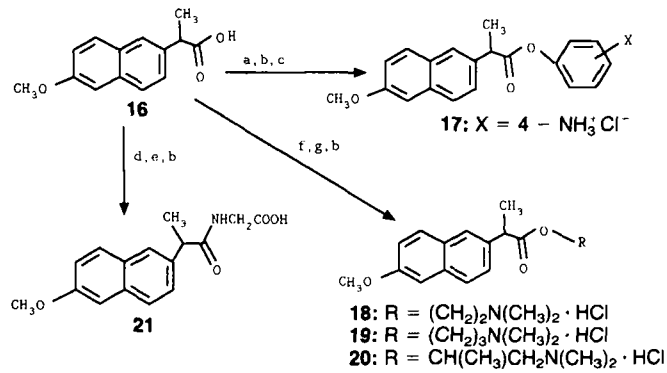
Scheme I—Synthesis of prodrugs **2–12**. Key: (**a**) *m*- or *p*-nitrophenol/DDC; (**b**) H_2/Pd-C; (**c**) HCl; (**d**) isopropenyl chloroformate DMAP/Et_3N; (**e**) BOC-$NHCH_2OH$; (**f**) $SOCl_2$; (**g**) ROH.



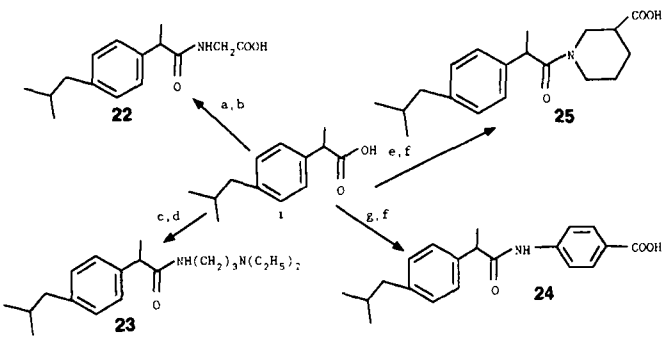
Scheme II—Synthesis of prodrugs **13–15**. Key: (**a**) $SOCl_2$; (**b**) $HOCH_2CH_2N(CH_3)CH_2C_6H_5$; (**c**) HCl (g); (**d**) 1-(2-hydroxyethyl)-4-benzylpiperazine; (**e**) H_2/Pd-C (50 °C); (**f**) H_2/Pd-C (25 °C).

h, the precipitated $Et_3N \cdot HCl$ was removed by filtration, and the solvent was evaporated to yield the free base. Ibuprofen (**1**; 5.0 g, 24.0 mmol) was dissolved in CH_3CN (150 mL) and added to a flask containing glycine benzyl ester. Dicyclohexylcarbodiimide (4.95 g, 24.0 mmol) was added to the solution, and stirring was continued overnight under anhydrous conditions. The precipitated dicyclohexylurea was filtered, and the solvent was evaporated under reduced pressure to afford a waxy semisolid. The residue was dissolved in ethanol (150 mL) and shaken on a Parr hydrogenator at 50 psi for 24 h in the presence of 0.5 g of 5% Pd/C. The catalyst was removed by filtration, and the solvent was evaporated to yield a solid. The solid was dissolved in 40% NaOH and extracted with diethyl ether (3 × 50 mL). The aqueous layer was separated, acidified with 6 M HCl, and cooled to yield a white solid. Recrystallization produced 2.2 g of white solid; IR (KBr): 1740 (C=O, acid), 1650 (C=O, amide) cm^{-1}; NMR ($CDCl_3$): δ 0.89 (d, 6 H, J = 6 Hz, $(CH_3)_2$), 1.49 (d, 3 H, J = 8 Hz, CH_3CH), 1.78 (m, 1 H, $(CH_3)_2CH$), 2.38 (d, 2 H, J = 6 Hz, CH_2Ar), 3.70 (q, 1 H, CH_3CH), 3.98 (d, 2 H, J = 6 Hz, CH_2COOH), 7.10 (m, 4 H, ArH).

4-[2-(4-Isobutylphenyl)propionamido]benzoic Acid (24)—A mixture of **1** (5.0 g, 24.0 mmol), *p*-aminobenzoic acid ethyl ester (4.0 g, 24.0 mmol), and dicyclohexylcarbodiimide (4.95 g, 24.0 mmol) in CH_3CN (150 mL) afforded 4.5 g of a waxy semisolid in a similar



Scheme III—Synthesis of prodrugs **17–21**. Key: (**a**) *p*-nitrophenol/DCC; (**b**) H_2/Pd-C; (**c**) HCl (g); (**d**) *N*-hydroxysuccinimide; (**e**) glycine benzyl ester/DMAP; (**f**) $SOCl_2$; (**g**) ROH.



Scheme IV—Synthesis of prodrugs **22–25**. Key: (**a**) glycine benzyl ester hydrochloride/Et_3N/DCC; (**b**) H_2/Pd-C; (**c**) $SOCl_2$; (**d**) $H_2NCH_2CH_2CH_2NEt_2$; (**e**) ethyl nipecotate/HOBt/DCC; (**f**) 25% NaOH; (**g**) ethyl *p*-aminobenzoate.

manner as described for the synthesis of **22**. The residue was dissolved in CH_2Cl_2 and washed with cold 5% NaOH (3 × 50 mL). The CH_2Cl_2 layer was separated, dried (Na_2SO_4), filtered, and evaporated under reduced pressure. The resultant solid was suspended in 25% NaOH (150 mL) and refluxed for 3 h. The solution was cooled, acidified with 6 M HCl, and refrigerated overnight to yield a white solid. Recrystallization gave 3.67 g of a white crystalline solid; IR (KBr): 1740 (C=O, acid), 1650 (C=O, amide) cm^{-1}; NMR ($CDCl_3$): δ 0.89 (d, 6 H, J = 6 Hz, $(CH_3)_2$), 1.51 (d, 3 H, J = 8 Hz, CH_3CH), 1.78 (m, 1 H, $(CH_3)_2CH$), 2.41 (d, 2 H, J = 6 Hz, CH_2Ar), 3.79 (q, 1 H, CH_3CH), 7.20 (q, 4 H, ArH), 7.9 (m, 5 H, NHArH).

1-[2-(4-Isobutylphenyl)propionyl]-3-piperidinecarboxylic Acid (25)—The synthesis of this compound was accomplished by the procedure reported by Thaisrivongs et al.[27] A mixture of **1** (3.5 g, 17.0 mmol), ethyl nipecotate (2.67 g, 17.0 mmol), and dicyclohexylcarbodiimide (3.5 g, 17.0 mmol) in CH_2Cl_2 (100 mL) was treated with 1-hydroxybenzotriazole hydrate (2.29 g, 17.0 mmol). The reaction mixture was stirred for 2 h at 0 °C and allowed to warm to room temperature overnight. The precipitated dicyclohexylurea was filtered, and the reaction mixture was washed successively with 1 M citric acid (3 × 50 mL), saturated NaCl (1 × 50 mL), and 1 M $NaHCO_3$ (3 × 50 mL). The organic phase was dried (Na_2SO_4), filtered, and evaporated to dryness under reduced pressure. The resulting oil was suspended in 25% NaOH (150 mL) and refluxed for 2 h. The reaction mixture was cooled and acidified with 6 M HCl to give a gummy mass. Trituration with acetone afforded a white solid which was recrystallized to yield 1.42 g of a crystalline solid; IR (KBr): 1740 (C=O, acid), 1650 (C=O, amide) cm^{-1}; NMR ($CDCl_3$): δ 0.89 (d, 6 H, J = 6 Hz, $(CH_3)_2$), 1.49 (d, 3 H, J = 6 Hz, CH_3CH), 1.78 (m, 1 H, $(CH_3)_2CH$), 1.95–2.10 (m, 6 H, $NCH_2CH_2CH_2$), 2.45 (d, 2 H, J = 8 Hz, CH_2Ar), 3.71 (q, 1 H, CH_3CH), 4.00 (m, 3 H, NCH_2 and $CHCOOH$), 7.15 (q, 4 H, ArH).

Anti-inflammatory Activity—Male Swiss-Webster mice (18–22 g) were fasted with access to water. Ibuprofen (**1**) and **16** were dissolved in saline with a few drops of 0.1 M NaOH, and the final pH values of the solutions were 8–8.5. All prodrugs tested were soluble in saline, and the pH of these solutions was adjusted so as to be identical to

those solutions containing 1 and 16. A 0.1-mL/20 g body wt of the parent drug, prodrug, or control was administered sc to mice 20 min prior to a 0.2-mL/20 g body wt ip injection of 0.6% acetic acid. The number of writhes for each mouse was noted for 20 min after the acetic acid injection. Assuming the number of writhes for the control to be 100%, the percentage writhing induced by acetic acid in the presence of parent drug and prodrugs was calculated[28,29] according to the following expression:

$$\% \text{ Writhing} = \frac{(\text{average number of writhes-drug})}{(\text{average number of writhes-control})} \times 100 \quad (1)$$

Gastrointestinal Toxicity—Male Sprague-Dawley rats (180–220 g, n = 6) were used in all experiments following reported procedures.[7,11,30,31] Ibuprofen (1), 16, and their respective glycine amides, 21 and 22, were suspended in normal saline using 2% Tween 80. The dimethylaminoethyl esters 5 and 18 were dissolved in normal saline containing 2% Tween 80. The compounds were administered orally by gavage in a volume of 10 mL/kg. Doses equivalent to 150 mg/kg of 1 and 40 mg/kg of 16 were used. The control animals were given 10 mL/kg of normal saline containing 2% Tween 80. Parent drugs or prodrugs were administered orally daily for 4 days. The animals were fasted for 8 h prior to dosing and for 4 h post dosing. Food was available at all other times, and free access to water was provided throughout the experiment. Four hours after the last dose, the animals were sacrificed using CO_2. The abdomen was opened at the midline, and the stomach and the first 3 cm of the duodenum were removed. The stomach was opened along the lesser curvature and washed with distilled H_2O. The mucus was wiped off, and the number of lesions was examined by means of a 2 × 2 binocular magnifier. All ulcers >0.5 mm were counted and recorded as average number of ulcers per compound (Table I).

Results and Discussion

Synthesis—Prodrugs of 1 and 16 were synthesized by standard procedures as shown in Schemes I–IV. The preparation of the aminoethyl ester 4 involved mixed anhydride formation using isopropenyl chloroformate.[22,23] Reaction of the mixed anhydride with BOC-aminoethanol,[24] followed by removal of the BOC-protecting group, gave the desired ester 4. The glycine amide 21 required activation of the carboxyl group of 16 through formation of an intermediate succinimido ester. Coupling of the active ester with glycine benzyl ester in the presence of DMAP, followed by hydrogenolysis of the O-benzyl group, afforded 21. The synthesis of the amide 25 was accomplished by activated ester formation between 1 and 1-hydroxybenzotriazole (HOBt). The physical properties of 1 and 16 prodrugs are given in Table II. All prodrugs of 16 were prepared from the (+)-enantiomer. Racemization occurred in the synthesis of naproxen esters 18 and 20. The naproxen esters 17 and 19 and the glycine amide 21 were optically active, showing the same sign of optical rotation as the parent acid. No attempt was made to determine the enantiomeric purity of these prodrugs.

Anti-inflammatory Activity—The anti-inflammatory activity of 1, 16, and a variety of their prodrugs, as measured by their ability to inhibit acetic acid-induced writhing in mice, is

Table II—Physical Properties of Ibuprofen and Naproxen Prodrugs

Compound	mp, °C (bp)[a]	Recrystallization		
		Yield, %[b]	Solvent[c]	Formula[d]
2	204–205	39	A	$C_{19}H_{24}ClNO_2$(C,H,N)
3	140–141	48	A	$C_{19}H_{24}ClNO_2$(C,H,N)
4	124–126	38	B	$C_{15}H_{24}ClNO_2$(C,H,N)
5	121–122 (116–117, 0.2 mmHg)	47	B	$C_{17}H_{28}ClNO_2$(C,H,N)
6	104–105 (122–125, 0.2 mmHg)	37	C	$C_{19}H_{32}ClNO_2$(C,H,N)
7	156–158 (133–135, 0.5 mmHg)	43	C	$C_{18}H_{30}ClNO_2$(C,H,N)
8	97–99 (155–162, 0.9 mmHg)	37	C	$C_{20}H_{34}ClNO_2$(C,H,N)
9	135–137 (116–117, 0.3 mmHg)	61	C	$C_{18}H_{30}ClNO_2$(C,H,N)
10	150–151 (127–128, 1.3 mmHg)	26	C	$C_{20}H_{34}ClNO_2$(C,H,N)
11	110–112 (155–162, 0.35 mmHg)	64	C	$C_{19}H_{30}ClNO_3$(C,H,N)
12	100 (145–152, 0.3 mmHg)	43	C	$C_{20}H_{32}ClNO_2$(C,H,N)
13	112–115 (175–179, 0.55 mmHg)	53	C	$C_{23}H_{32}ClNO_2$(C,H,N)
14	116–117	55	B	$C_{16}H_{26}ClNO_2$(C,H,N)
15	224 (dec)	51	D	$C_{18}H_{32}Cl_2N_2O_2$(H,N)[e]
17[f]	241–242	38	E	$C_{20}H_{20}ClNO_3$(C,H,N)
18[g]	120–123 (188–192, 1.6 mmHg)	47	F	$C_{18}H_{24}ClNO_3$(C,H,N)
19[h]	143–144	52	F	$C_{19}H_{26}ClNO_3$(C,H,N)
20[g]	143–145 (185–195, 1.5 mmHg)	48	F	$C_{19}H_{26}ClNO_3$(C,H,N)
21	132–134	40	G	$C_{16}H_{17}NO_4$(C,H,N)
22	79–81	35	G	$C_{15}H_{21}NO_3$(C,H,N)
23	(170–175, 0.5 mmHg)	28		$C_{20}H_{34}N_2O$(C,H,N)
24	240–241	47	G	$C_{20}H_{33}NO_3$(C,H,N)
25	119–121	26	H	$C_{19}H_{27}NO_3$(C,H,N)

[a] Boiling point of free base. [b] Yields are calculated on the recrystallized hydrochloride salts. [c] A = EtOH; B = EtOAc; C = EtOAc:Et_2O; D = 95% EtOH; E = EtOH:Et_2O; F = 95% EtOH:Et_2O; G = EtOH:H_2O; H = CH_3COCH_3:H_2O. [d] All new compounds were analyzed for the elements shown in parentheses and were within ±0.4% of the calculated value, unless otherwise specified. [e] C: calc, 58.30; found, 58.89. [f] $[\alpha]_D^{25°}$ + 48° (c 1, CH_3OH). [g] Racemized during synthesis. [h] $[\alpha]_D^{25°}$ + 30° (c 1, CH_3OH). [i] $[\alpha]_D^{25°}$ + 21.8° (c 1, CH_3OH).

Table III—Anti-Inflammatory Activity in Mice[a]

Pretreatment	% Writhes[b]
Saline	100.0 ± 8.0
Ibuprofen (1)	32.5 ± 8.0
3	13.5 ± 5.5
5	19.2 ± 9.8
7	11.2 ± 7.2[c]
9	15.3 ± 10.2
15	12.2 ± 6.3[c]
22	27.2 ± 10.8
Naproxen (16)	15.9 ± 0.12
17	11.3 ± 3.50
18	26.9 ± 2.90
19	3.7 ± 1.80[c]
20	5.4 ± 0.60[c]
21	22.8 ± 9.9

[a] Doses administered: Ibuprofen (1; 44 mg/kg, 0.213 mmol/kg); ibuprofen (1) prodrugs 3, 5, 7, 9, 15, and 22 (0.213 mmol/kg); naproxen (16; 22 mg, 0.096 mmol/kg); naproxen (16) prodrugs 17, 18, 19, 20, 21 (0.096 mmol/kg). [b] All values are mean ± SD (n = 6). [c] $p < 0.01$.

given in Table III. The prodrugs which were evaluated for anti-inflammatory activity were selected on the basis of chemical structure, relative lipophilicity, chemical stability, and rates of enzymatic hydrolysis[32] in human serum. The saline control was considered to exhibit 100% writhing, and the protection afforded by the parent drugs and prodrugs was calculated on a percentage basis. A one-way ANOVA was run on the data to determine which of the prodrugs were significantly different from the parent drugs. Compounds 7, 15, 19, and 20 exhibited significantly better activity than the parent compounds ($p < 0.01$). All other prodrugs were not significantly more active than the parent compounds. Interestingly, the racemic ester 20 exhibited significantly better anti-inflammatory activity than the parent NSAID 16 having the *S* configuration. The inhibition of prostaglandin synthesis by 2-arylpropionic acids, such as 1 and 16, has been shown to reside almost exclusively in the *S*-enantiomer. Although a unidirectional chiral inversion has been shown to occur in humans with 1, 16 apparently does not undergo this transformation in humans or rats.[33]

Ulcerogenic Activity—The average number of ulcers formed in the gastric mucosa following oral administration of 1, 16, and prodrugs 5, 18, 21, and 22 is given in Table I. The dimethylaminoethyl esters 5 and 18 and the glycine amides 21 and 22 were selected for evaluation. The esters 5 and 18 were tested since they are stable in simulated gastric fluid, exhibit a rapid breakdown to the parent compounds in human serum,[32] and exhibit anti-inflammatory activity. In contrast, the amides 21 and 22 are hydrolyzed considerably slower in human serum ($t_{1/2}$ = 390 and 300 min, respectively[32]). These amides exhibit anti-inflammatory activity, although not significantly better than the parent NSAID. The ulcerogenic activities of these amide prodrugs were of interest since they contain a free carboxylic acid group. All prodrugs, except the glycine amide 21, were significantly less irritating to the gastric mucosa than the parent NSAID (one-way ANOVA, $p < 0.01$). Although the intestinal mucosa was also observed, no detectable ulcers were noted.

Conclusions

Several prodrugs of 1 and 16 were equal or better inhibitors of acetic acid-induced writhing in mice, as compared with the parent NSAID (Table III). As previously stated, the direct contact mechanism in gastric ulceration is a combination of the local irritation produced by the free carboxylic acid group of the NSAID and the local inhibition of the cytoprotective actions of prostaglandins on the gastric mucosa. As the prodrugs 5, 18, 21, and 22 remain intact over a 4-h time period in simulated gastric fluid, it can be assumed that they are absorbed intact.[32] Hence, it appears that gastrointestinal irritation produced by these compounds probably arises from systemic inhibition of prostaglandin synthesis, due to the conversion of the prodrug to 1 or 16 in the blood following absorption of the prodrug from the gastrointestinal tract.

Thus, the results of this study strongly support the existence of a direct contact mechanism and a systemic action of gastrointestinal irritation. Furthermore, the results obtained with the ester prodrugs demonstrate that the direct contact mechanism is more critical than the systemic action in gastrointestinal irritation. Additionally, within the direct contact mechanism, inhibition of prostaglandin synthesis is more significant than the effect of the free carboxylic acid group of the NSAID. This hypothesis is supported by the fact that the glycine amides 21 and 22 produce a low degree of gastrointestinal irritation, even though they contain a free carboxylic acid group.

In summary, ester and amide prodrugs of 1 and 16 represent a potentially useful method to decrease gastrointestinal side effects without altering the pharmacological profile of the parent compounds.

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