Pergamon

Tetrahedron, Vol. 53, No. 32, pp. 10953-10970, 1997

0040-4020/97 $17.00 + 0.00

PII: S0040-4020(97)00357-8

Convergent Catalytic Asymmetric Synthesis of Camptothecin Analog GI147211C[1]

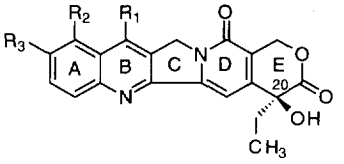
Francis G. Fang,* Donald D. Bankston,[2] Edward M. Huie, M. Ross Johnson,[3]Myung-Chol Kang,[3] Craig S. LeHoullier, George C. Lewis,[4] Thomas C. Lovelace, Melissa W. Lowery, Darryl L. McDougald, Clive A. Meerholz, John J. Partridge, Matthew J. Sharp, and Shiping Xie

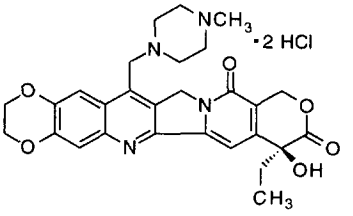
Chemical Development Department, Glaxo Wellcome Inc.,
Research Triangle Park, North Carolina 27709

Abstract: The topoisomerase I inhibitor GI147211C (**4**) was discovered at Glaxo Wellcome and shown to have promising anti-cancer properties. In order to fully assess the clinical potential of **4**, an improved synthesis of the drug substance was required. Herein is described a convergent catalytic asymmetric synthesis of **4** which utilizes as key steps , two Heck reactions, a Sharpless asymmetric dihydroxylation reaction, and a Mitsunobu reaction. A 2-chloroquinoline is shown to be a viable substrate for the final Heck reaction to generate the camptothecin nucleus. © 1997 Elsevier Science Ltd.

The structure and novel mechanism of action as a topoisomerase I inhibitor have combined to make the natural product camptothecin (**1**) one of the more compelling targets for new cancer chemotherapy over the past three decades.[5] And yet, presumably due to its poor water-solubility and related poor pharmacodynamic properties, the pentacyclic array embodied by the natural product **1**, is itself apparently inadequate to be a useful medicine. With the recent FDA approval of topotecan (**2**) and irinotecan (**3**), water-soluble analogs of **1** are now established as useful additions to the anti-cancer armamentarium.[6,7] Both **2** and **3** are the products of semi-synthetic programs starting from the naturally occurring 20(S)-camptothecin (**1**).[8,9] Two limitations of this strategy for analog development are: (i) Supply of drug substance is dependent on consistent availability of 20(S)-camptothecin (**1**) and (ii) Current methodology for functionalizing **1** restricts the range of analog structures which can be obtained directly from the natural product. Our efforts to develop a camptothecin analog at Glaxo Wellcome have focused on using total synthesis to provide access to structures, not available by derivatization of the natural product **1**. This strategy would hopefully allow for a more comprehensive assessment of the key structural parameters required for optimum biological efficacy. A result of these efforts was the discovery of the "fully synthetic" camptothecin analog GI147211C (**4**), a potent and novel water-soluble topoisomerase I inhibitor which is currently undergoing phase II clinical evaluation for the treatment of a variety of tumor types.[10]

Figure 1

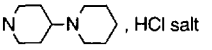




1, 20(S)-camptothecin, $R_1 = R_2 = R_3 = H$,

4 GI147211C

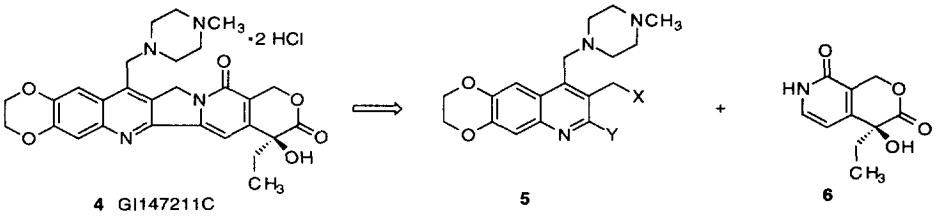
2, topotecan, $R_1 = H$, $R_2 = CH_2NMe_2$, $R_3 = OH$, HCl salt

3, irinotecan, $R_1 = Et$, $R_2 = H$, $R_3 = OCO$  , HCl salt

While a research program which specifically targets "fully synthetic" analogs (i.e. analogs which are not found in nature, nor are derivable in a direct fashion from the natural product) potentially offers a greater diversity of analog structures (than a semi-synthetic program), it is also by definition completely reliant on total synthesis for the discovery and further development of such a drug candidate. Once GI147211C (**4**) was identified as a promising candidate for further studies, the need for a practical synthesis became urgent. The medicinal chemistry synthesis of **4** had relied on the Wall CDE ring fragment[11] for its preparation. Although this approach allowed for the rapid construction of a range of analogs, the long linear nature, need for a late-stage resolution, and low-yield piperazine installation made it less than desirable for further development.[10] A second synthesis of **4** recently reported by Curran et al.,[12] applies our recently reported catalytic asymmetric DE-ring synthesis[13] to the preparation of a halo-functionalized DE-ring and combines this with a cascade radical cyclization to generate the BC-rings in a single step, albeit with limited regiocontrol (3:2 desired:undesired in the formation of the B-ring of **4**). A new total synthesis of **4** was clearly required.

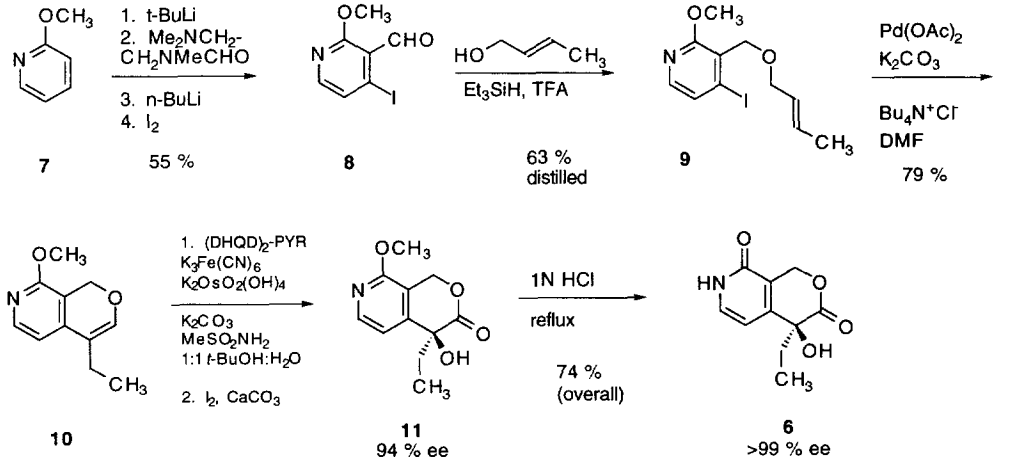
Over the past twenty-five years, numerous studies have been reported dealing with the synthesis of camptothecin (**1**).[14,15] For our purposes, the most promising of these appeared to be the convergent asymmetric synthesis recently reported by Comins *et al* which joins the AB and DE-ring (**6**) fragments to form **1** via a sequential pyridone N-alkylation and Heck reaction.[15e, 16] At the time our work began on applying this strategy to the synthesis of **4** (Scheme 1), there were three significant issues. First, use of stoichiometric amounts of an expensive chiral auxiliary ((-)-8-phenylmenthol) in the Comins synthesis to selectively form the 20(S)-stereogenic center limits availability of the DE-ring fragment **6**. Second, there would be a need for an efficient preparation of the "AB" ring portion of **4** (i.e. **5**) along with group Y needed for the intramolecular Heck reaction. And third, the N-alkylation of **6** with **5** would involve a leaving group (X) located δ to a nitrogen atom raising the issue of stability of intermediate **5**. We have recently reported the first catalytic asymmetric synthesis of the camptothecin DE-ring (**6**) which effectively deals with the first concern.[13] Herein we report: (i) a short synthesis of viable forms of **5** (X=OH, Y=Cl, Br, I), (ii) an efficient N-alkylation of **6** with fragment **5**, and (iii) the first palladium mediated intramolecular Heck reaction of a 2-chloroquinoline.

Scheme 1



The reactions to prepare the DE-ring fragment are described with full experimental detail in our previous communication[13] and are outlined in **Scheme 2** below. Key reactions include a tandem Heck reaction[17] / olefin isomerization process and a highly enantioselective Sharpless asymmetric dihydroxylation (AD) reaction using the pyrimidine based catalyst[18] to establish the C.20 stereocenter. A final hydrolysis of the methoxypyridine **11** in dilute hydrochloric acid directly precipitates the enantiomerically pure DE-ring **6** as a crystalline solid. Some aspects of these reactions continue to be developed and will be further disclosed separately.[19]

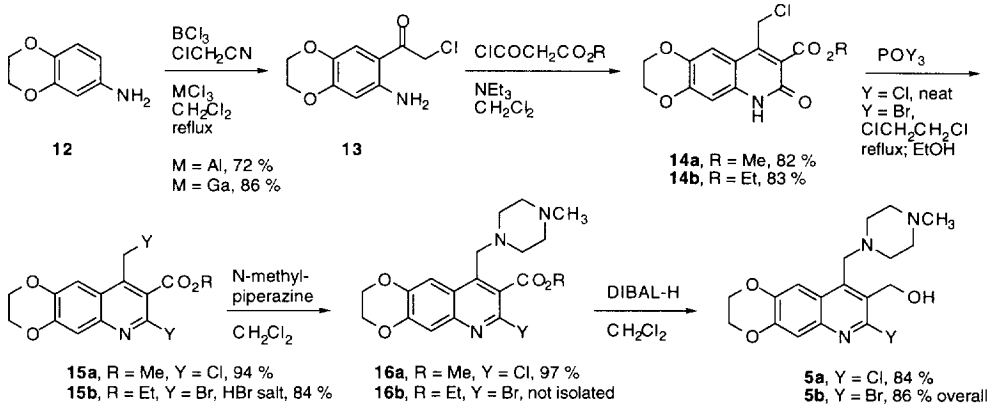
Scheme 2



Synthesis of the requisite fragment **5** needed for coupling with fragment **6** is set forth in **Scheme 3** below. The first step, boron trichloride and aluminum trichloride mediated Friedel-Crafts acylation of benzodioxane-6-amine (**12**) with chloroacetonitrile has previously been reported to give less than 35 % of product **13**.[10] Significantly improved results were obtained by inverting the order of addition of the aniline and

the boron trichloride (i.e. adding the aniline to the boron trichloride) to give 72 % (M = Al) of **13**. Alternatively, following the "normal" order of addition (adding the boron trichloride to the aniline) but including a minimum 2 hour aging period at room temperature prior to adding the electrophile (chloroacetonitrile) and second Lewis acid (aluminum or gallium trichloride) provided 86 % (M = Ga) of **13**. In addition to offering a modestly more efficient process, the use of gallium trichloride[20] facilitated the work-up by avoiding the formation of emulsions seen when using aluminum trichloride. This method for ortho-acylation of anilines, first reported by Sugasawa et al.,[21] requires the use of two different Lewis acids, the boron trichloride to coordinate to the aniline nitrogen, and a second Lewis acid (aluminum trichloride or gallium trichloride) to activate the nitrile. Together, these results underscore the importance of forming a one-to-one complex of the aniline with boron trichloride in order to obtain high yields of product.

Scheme 3



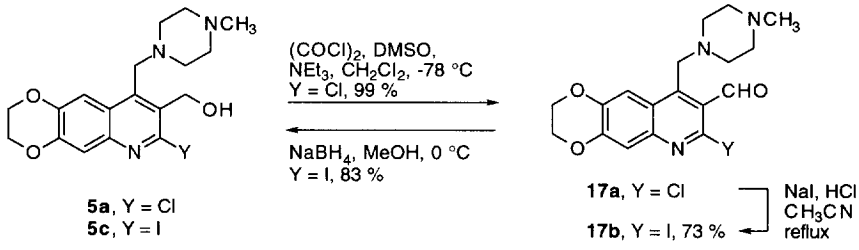
Annulation of the acyl aniline **13** to the quinolone **14** is achieved by N-acylation with methyl or ethyl malonyl chloride followed by based-induced aldol condensation.[22] Both the acylation and aldolization processes are mediated by triethylamine, however, the amine must be added in two separate charges (1 eq. during the acylation and 1 eq after the acylation is complete) in order to efficiently accomplish the overall transformation.

We initially considered a bromo- or iodo- quinoline a necessary prerequisite for a fully actuatable left-hand component. This assumption was based on the work of Comins, who had shown that either a bromo- or iodo-quinoline underwent the final Heck cyclization to yield camptothecin (**1**).[15e] The intent was to form the iodoquinoline from the chloroquinoline by halide exchange. Thus, treatment of quinolone **14a** with phosphorous oxychloride provided after quenching into ice-water, a 94 % yield of the chloroquinoline **15a**. The reaction of **14b** with commercially available phosphorous oxybromide in refluxing 1,2-dichloroethane, followed

by quenching with ethanol precipitated the bromoquinoline as the hydrobromide salt, **15b**, in 84 % yield. The use of ethanol served two purposes here: (i) to quench any trace of acid bromide formed during the reaction, and (ii) to provide a highly crystalline and easily isolable hydrobromide salt.

Treatment of the chloromethylchloroquinoline **15a** with N-methylpiperazine (2 eq.) in dichloromethane yielded the adduct **16a** in 97 % yield. Reduction of this methyl ester gave the chloroquinoline alcohol left-hand fragment, **5a**. Similarly, treatment of **15b** with N-methylpiperazine (1.25 eq.) and triethylamine in dichloromethane afforded the corresponding bromoquinoline, **16b**, which was not isolated but carried directly into the reduction step. Treatment of **16b** with diisobutylaluminum hydride in dichloromethane produced the bromoquinoline **5b** in 86 % overall yield (for two steps). With both the chloro- and bromo-quinolines in hand, we turned to incorporation of the iodide to complete the series. Compound **5a** was itself too inactive to allow this exchange process. In order to improve the situation, **5a**, was converted to an aldehyde by oxidation (see **Scheme 4**) and then subjected to standard conditions for the exchange of a chloride and iodide. The use of more than 2 equivalents of hydrochloric acid was necessary to induce this transformation, presumably to tie-up the two basic piperazine nitrogens. Sodium borohydride reduction of aldehyde **17b** in methanol provided the iodoquinoline fragment **5c** in 83 % yield.

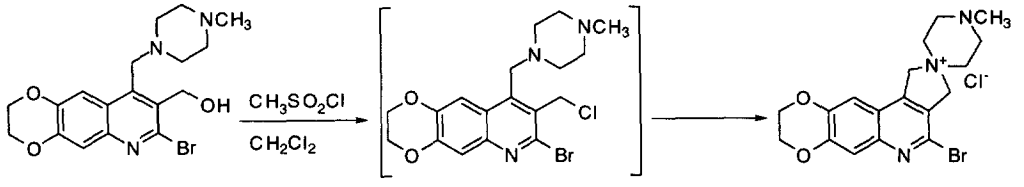
Scheme 4



With flexible access to the required left hand fragments, we could now study the feasibility of coupling these pieces with the DE-ring, **6**. Following on the precedent of Comins, this would involve transforming the alcohol moiety to a halide leaving group followed by base-mediated displacement with **6**.[15e,16] Owing to the fact that **5** possesses a tertiary piperazine nitrogen δ to the halide, we suspected that this material might be prone to quaternization (*vide infra*) and thus be difficult to work with as an isolable intermediate. The experiment shown below (see **Scheme 5**) helped to confirm this. Thus, the alcohol **5b** was stirred with methanesulfonyl chloride in dichloromethane at room temperature. No amine was added since the piperazine nitrogen could serve as an internal base. In fact protonation of this nitrogen atom might discourage the aforementioned quaternization process. Over a 2 hour period, the initially streaky tlc plate converged to mainly one less polar

new spot. A brief aqueous work-up and concentration of solvent left the chloride, **18**. Upon standing, **18** converts to the quaternary salt **19**. Attempts to use **19** in condensation reactions with **6** were unsuccessful.

Scheme 5



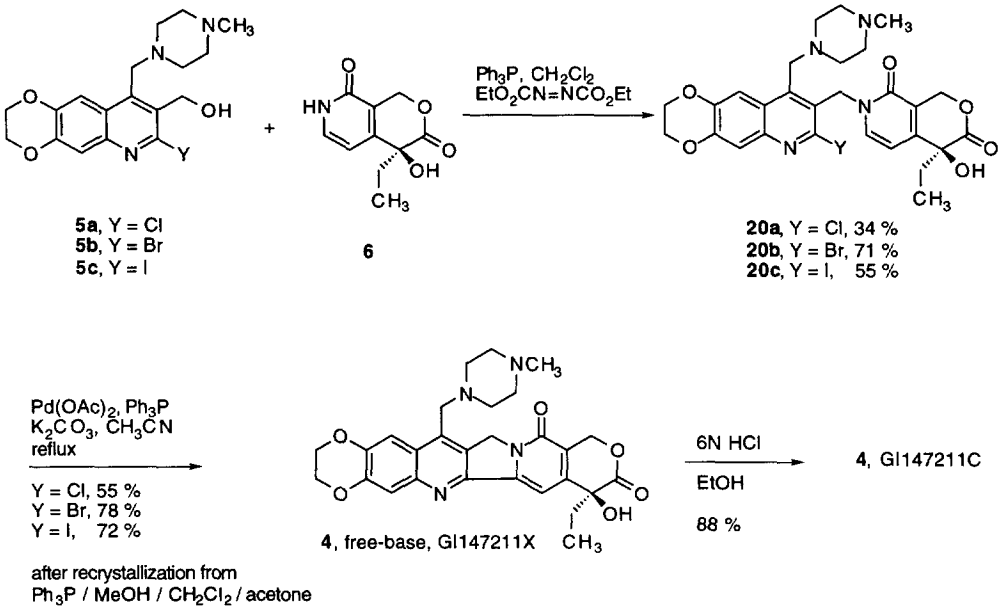
5b **18** **19**

Hoping to minimize the formation of **19**, we turned to an *in situ* method (Mitsunobu conditions) for activation of the alcohol **5**.[23-24] A suspension of **5b** and **6** in dichloromethane and triphenylphosphine was treated with diethylazodicarboxylate at room temperature. As the addition progressed, the reaction warmed to reflux and the reaction components went into solution. Upon completion of the addition, and after removal of trace undissolved solids, the product **20b** precipitated in 71 % yield upon addition of methyl t-butyl ether. Similar conditions yielded the corresponding chloro- and iodoquinolines **20a** and **20c** in 34 % and 55 % (unoptimized yields) respectively (see **Scheme 6**).

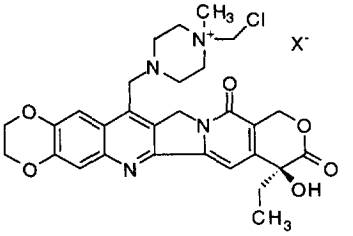
The key Heck cyclization was carried out using palladium(II) acetate, triphenylphosphine, potassium carbonate, in acetonitrile at reflux. There were several differences from the original conditions reported by Comins: (i) No phase transfer catalyst was found to be necessary for these substrates, (ii) triphenylphosphine and potassium carbonate (powdered) significantly improved the rate and conversion for the process, and (iii) Use of acetonitrile allowed the product to precipitate from the reaction mixture as it formed. Under these conditions, we found that all three haloquinolines, bromo-, iodo-, and even chloro- smoothly underwent the cyclization reaction to provide the final product **4** (free-base) in the indicated yields. The cyclization of the chloroquinoline was unexpected. To our knowledge, few examples exist for such a process and these usually involve forcing conditions.[25,26] Two recrystallizations of the product from dichloromethane, methanol, and acetone containing triphenylphosphine served to remove traces of palladium from the product to give metal-free drug substance, **4**, as the free-base.[27]

A final observation during the exercise to remove palladium from the drug substance serves to act as a caution to the use of halogenated solvents, phosphines, and amines together. In an attempt to expedite the clean-up of drug substance, tri-n-butyl phosphine substituted for triphenylphosphine as a ligand for palladium. A trace impurity, previously seen at <0.1 %, was now detected at ca. 3 %. Isolation of this material and identification by mass spec, ^{1}H-NMR, and ^{13}C-NMR revealed it to be the chloromethylammonium salt at the N-methylpiperazine nitrogen (i.e. **21**).[28] Formally this process could be considered a Menschutkin reaction,

Scheme 6



although typically the quaternization of amines with chlorides is conducted under extreme conditions (temperature and pressure).[29] Perhaps the more reactive tributylphosphine accelerated a process which might involve phosphine displacement of chloride from dichloromethane, followed by amine displacement of phosphine from this intermediate. This process may have practical application elsewhere as a means of selectively catalyzing ammonium salt formation.



21

The final step of the synthesis, conversion of the free-base **4**, GI147211X to the dihydrochloride salt **4**, GI147211C was carried out in a straightforward fashion. The free-base is dissolved in warm 6N hydrochloric

acid, filtered through a membrane filter, and diluted with ethanol to precipitate the final drug substance, **4**, as a yellow powder in 88 % yield.

In summary, we have demonstrated a process for the large-scale preparation of the fully synthetic camptothecin analog, GI147211C, **4**. All steps in this sequence yield crystalline or distillable intermediates and can be carried out on kilogram scale without chromatographic purifications. In so doing, we have described: (i) Practical access to both the left and right-hand fragments, (ii) efficient methods for connecting these to form the final drug substance, and (iii) the first palladium mediated Heck reaction of a 2-chloroquinoline. These results should find practical application in the large-scale preparation of other fully synthetic camptothecin analogs which are useful as potential anti-cancer agents.

Experimental Procedures:

Preparation of 1-(7-amino-2,3-dihydro-benzo[1,4]dioxin-6-yl)-2-chloro-ethanone, 13:

A solution of 1 M boron trichloride in dichloromethane (4 L, 4.00 mol) was cooled to -20 °C. To this was added a solution of 1,4-benzodioxane-6-amine (500 g, 3.31 mol) in 250 mL of dichloromethane (DCM) over 30 min. The reaction mixture warmed to 10 °C during the addition and was subsequently re-cooled back to -10 °C. Chloroacetonitrile (250 mL, 3.95 mol) was added over 5 min, followed by aluminum chloride (485 g, 3.64 mol). The resultant dark solution was heated at reflux for 24 h. After being cooled to room temperature, the mixture was transferred to 2-20 L separatory funnels, each containing 10 L of water. After being stirred for 2.5 h, the organic layer was separated and the aqueous layer was extracted with DCM (4 x 4 L). The combined organic layers were washed with brine (4 L), dried over anhydrous sodium sulfate and concentrated *in vacuo*. The resultant brown slurry was successively treated with 600 mL of DCM and 2 L of hexane. The mixture was stirred at 0 °C for 30 min. The precipitate was collected by filtration, washed with 1 L of hexane and dried *in vacuo* at RT to afford 544 g (72%) of product **13** as a yellow powder. Reverse phase HPLC (Spherisorb ODS-25 micron, 1:1 MeCN-H_2O) indicated a purity of 91%. Recrystallization from dichloromethane provided an analytical sample as a yellow crystalline solid: mp 130 °C (dec.). ^1H NMR ($CDCl_3$, 200 MHz) δ 4.24 (m, 2H), 4.32 (m, 2H), 4.59 (s, 2H), 6.21 (s, 1H), 6.23 (br. s, 2H), 7.29 (s, 1H). IR (KBr) 3450, 3375, 2900, 1650, 1580, 1321 cm^{-1}. Elemental analysis: Calculated for $C_{10}H_{10}ClNO_3$: C 52.76, H 4.43, N 6.15. Found: C 52.62, H 4.42, N 6.12.

Preparation of 1-(7-amino-2,3-dihydro-benzo[1,4]dioxin-6-yl)-2-chloro-ethanone, 13 (using gallium chloride):

A solution of 1,4-benzodioxan-6-amine (160 g, 1.06 mol) in dichloromethane (DCM, 2.4 L) is cooled in an ice bath to less than 10 °C. Boron trichloride gas (150 g, 1.27 mol, 1.2 eq) is added over 0.75 hour. The resulting brown slurry is then stirred overnight at room temperature. The slurry is then cooled in an ice bath to 10 °C and chloroacetonitrile (94 mL, 1.48 mol, 1.4 eq) is added in one portion. Gallium chloride (205 g, 1.16 mol, 1.1 eq)

is dissolved in DCM (310 mL) and added to the reaction over 0.5 h. The resulting brown slurry is then heated at reflux overnight (13- 24 h). The brown solution is then cooled to rt and poured into a stirred mixture of DCM (6.4 L)) and water (6.4 L). The mixture is stirred for 1.5 to 2 h to allow the solids to dissolve. The layers are then separated and the aqueous phase is extracted with DCM (2 L). The organic layers are combined and concentrated under vacuum (about 100 mm Hg) to about 320 mL. (Crystallization occurs during the concentration.) Heptane (640 mL) is added over 1 hour and the slurry stirred overnight at room temperature. The slurry is filtered, washing the cake with heptane (200 mL). The cake is then dried overnight at 40 °C under high vacuum (about 1 mm Hg) to give 206 g (86%) of 1-(7-amino-2,3-dihydro-benzo[1,4]dioxin-6-yl)-2-chloro-ethanone **13** as a yellow solid.

Preparation of 9-chloromethyl-7-oxo-2,3,6,7-tetrahydro[1,4]dioxino[2,3-g]quinoline-8-carboxylic acid methyl ester, 14a

To a mixture of chloro ketone **13** (595 g, 2.61 mol) and triethylamine (474 mL, 3.40 mol) in acetonitrile (3.5 L) at 0 °C was added methyl malonyl chloride (364 mL, 3.40 mol) over 35 min. The cooling bath was removed and the mixture was stirred for 5 h. To the resultant slurry was added 25% sodium methoxide in methanol (596 mL, 2.61 mol) over 10 min. After being stirred at room temperature for 2 h, the now very thick yellow slurry was diluted with water (3.5 L). The mixture was filtered and the filter cake was washed with water (3 L). The yellow solid was dried in vacuo at 60 °C to afford 665 g (82%) of product **14a** as a yellow solid. This crude product was used for the next step without further purification. Recrystallization from MeOH-DMSO (1:1) gave an analytical sample: mp >300 °C (dec.). ^{1}H NMR (DMSO-d_6, 400 MHz) δ 3.85 (s, 3H), 4.32 (s, 2H), 4.36 (s, 2H), 4.83 (s, 2H), 6.83 (s, 1H), 7.40 (s, 2H), 12.0 (s, 1H). IR (KBr) 2980, 1751, 1655, 1505, 1210 cm^{-1}. Elemental analysis: Calculated for $C_{10}H_{10}ClNO_3$: C 54.29, H 3.91, N 4.52. Found: C 53.68, H 3.84, N 4.48. HRMS (EI+): Calculated for $C_{10}H_{10}ClNO_3$: 309.0404. Found: 309.0405.

Preparation of 9-chloromethyl-7-oxo-2,3,6,7-tetrahydro[1,4]dioxino[2,3-g]quinoline-8-carboxylic acid ethyl ester, 14b

To a mixture of **13** (230 g, 1.01 mol) in acetonitrile (1.5 L) and triethylamine (197 mL, 1.41 mol, 1.4 eq) is added ethyl malonyl chloride (180 mL, 1.41 mol, 1.4 eq) over 0.5 h while keeping the reaction temperature less than 30 °C with an ice bath. After the addition is complete the reaction is stirred 1.5 h at room temperature. A second charge of triethylamine (140 mL, 1.01 mol, 1.0 eq) is added and the reaction is stirred for 4.5 hours. Water (2.1 L) is added slowly followed by conc. hydrochloric acid (115 mL). The slurry is stirred overnight at rt, filtered, and the solid washed with water (460 mL). The solid is then dried under high vacuum (about 1 mm Hg) at 40 °C to give 270 g (83%) 9-chloromethyl-7-oxo-2,3,6,7-tetrahydro[1,4]dioxino[2,3-g]quinoline-8-carboxylic acid ethyl ester as a yellow solid. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ 1.29 (t, J = 7.1 Hz, 3H), 4.3 (m, 6H), 4.8 (s, 2H), 6.8 (s, 1H), 7.39 (s, 1H). HRMS (EI$^+$): calc for $C_{15}H_{14}NO_5Cl$: 323.0561, Found: 323.0556.

Preparation of 7-chloro-9-chloromethyl-2,3-dihydro[1,4]dioxino[2,3-g]quinoline-8-carboxylic acid methyl ester, 15a

A mixture of chloro pyridone **14a** (360 g, 1.16 mol) in 1.8 kg of phosphorus oxychloride was heated at reflux for 20 h. The reaction mixture was allowed to cool to room temperature and slowly transferred to a 25-L separatory funnel containing 18 L of ice water. After being stirred vigorously for 1.5 h, the precipitate was collected by filtration, washed with 3 L of water and dried *in vacuo* at 50 °C to afford 358 g (94%) of product **15a** as a dark crystalline solid: mp 130-132 °C. ^{1}H NMR ($CDCl_3$, 200 MHz) δ 4.05 (s, 3H), 4.42 (s, 4H), 4.81 (s, 2H), 7.50 (s, 2H). IR (KBr) 2980, 1738, 1590, 1520, 1230 cm^{-1}. Elemental analysis: Calculated for $C_{14}H_{11}Cl_2NO_4$: C 51.24, H 3.38, N 4.27. Found: C 51.10, H 3.34, N 4.33.

Preparation of 7-bromo-9-bromomethyl-2,3-dihydro[1,4]dioxino[2,3-g]quinoline-8-carboxylic acid ethyl ester hydrobromic acid salt, 15b

To a mixture of **14b** (100 g, 0.31 mol) in 1,2-dichloroethane (DCE, 500 mL) is added a solution of phosphorus oxybromide (212 g, 0.62 mol) in DCE (250 mL). The resultant mixture is heated at reflux for 5.5 h, and then cooled to <15 °C. Ethanol (434 mL, 20.0 eq) is added while keeping the temperature <20 °C. The mixture is then allowed to warm to room temperature and stirred overnight. The mixture is filtered and the solid is washed with DCE (120 mL). The solid is dried under high vacuum (about 1 mm Hg) at rt overnight to yield 133.4 g (84 %) of **15b** as a yellow solid. ^{1}H NMR ($CDCl_3$, 200 MHz) δ 1.44 (t, J = 7.1 Hz, 3H), 4.43 (s, 4H), 4.51 (q, J = 7.1 Hz), 4.82 (s, 3H), 7.38 (s, 1H), 7.60 (s, 1H): HRMS (EI$^+$): calc for $C_{15}H_{13}Br_2NO_4$: 428.9211, Found: 428.9238.

Preparation of 7-chloro-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinoline-8-carboxylic acid methyl ester, 16a

A solution of dichloride **15a** (340 g, 1.04 mol) in 2 L of DCM is treated with N-methylpiperazine (237 mL, 2.14 mol) over 10 min. The solution was stirred at room temperature for 15 h. The reaction mixture was poured into 3 L of water and the organic layer was separated. The aqueous layer was extracted with DCM (3 x 2 L). The combined organic layers were washed with brine (2 L), dried over anhydrous sodium sulfate and concentrated *in vacuo*. The resultant thick brown slurry was successively treated with 300 mL of DCM and 1.5 L of hexane. The mixture was swirled at 0 °C for 30 min. The precipitate was collected by filtration and the cake washed with hexane (1 L) and dried *in vacuo* at rt to afford 392 g (97%) of product **16a** as a yellow powder: mp 142-143 °C. ^{1}H NMR ($CDCl_3$, 200 MHz) δ 2.28 (s, 3H), 2.38-2.49 (m, 8H), 3.81 (s, 2H), 3.96 (s, 3H), 4.59 (s, 2H), 4.40 (s, 4H), 7.45 (s, 1H), 7.65 (s, 1H). IR (KBr) 2940, 2794, 1657, 1506, 1433, 1240, 1107 cm^{-1}. Elemental analysis: Calculated for $C_{19}H_{22}ClN_3O_4$: C 58.24, H 5.66, N 10.72. Found: C 58.08, H 5.72, N 10.63.

Preparation of [7-chloro-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-yl]-methanol, 5a

To a solution of chloro ester **5** (194 g, 495 mol) in 1 L of DCM was added 1 M diisobutylaluminum hydride in DCM (2.00 L, 2.00 mol) over 15 min. The solution warmed to reflux during the addition. The reaction was allowed to cool to room temperature and stirred for 4 h. The reaction mixture was transferred to a 15-L separatory funnel containing 5 L of a saturated solution of Rochelle's salt. After being stirred for 2.5 h, the organic layer was separated and the aqueous layer was extracted with DCM (3 x 2.5 L). The combined organic layers were washed with brine (3 L), dried over anhydrous sodium sulfate and concentrated *in vacuo*. The resultant thick brown slurry was successively treated with DCM (0.5 L) and hexane (1 L). The mixture was swirled at 0 °C for 30 min. The precipitate was collected by filtration, washed with hexane (1 L) and dried *in vacuo* at 30 °C to afford 124 g (84%) of product **5a** as a yellow crystalline solid: mp 178-180 °C. ^{1}H NMR ($CDCl_3$, 200 MHz) δ 2.26 (s, 3H), 2.63 (br. s, 4H), 4.00 (s, 2H). 4.39(s, 4H), 4.93 (s, 2H), 6.10 (br. s, 1H), 7.46 (s, 1H), 7.51 (s, 1H). IR (KBr) 3200, 2965, 2848, 1562, 1504, 1299, 1019 cm^{-1}. Elemental analysis: Calculated for $C_{18}H_{22}ClN_3O_3$: C 59.42, H 6.09, N 11.55. Found: C 59.41, H 6.12, N 11.46.

Preparation of [7-bromo-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-yl]-methanol, 5b

To a mixture of **15b** (10 g, 19.6 mmol) in dichloromethane (DCM, 100 mL) is added triethylamine (5.4 mL. 39.2 mmol) followed by N-methylpiperazine (2.79 mL, 25.5 mmol). After stirring 30 min, water (50 mL) is added and the mixture is stirred 10 min. The layers are separated and the aqueous phase is extracted with DCM (50 mL). The organic phases are combined and washed with water (2x50 mL). The organic solution is then concentrated to an oil. The oil is diluted with DCM (50 mL) and stirred while diisobutylaluminum hydride (1.0 M in dichloromethane, 53 mL. 53 mmol) is added slowly while keeping the reaction temperature below 30 °C using an ice bath. The reaction is stirred for 10 min and then slowly poured into an aqueous solution (75 mL) saturated with potassium sodium tartrate tetrahydrate (Rochelle's salt). The temperature is kept <30 °C with an ice bath during the addition. The mixture gels and is stirred overnight at room temperature. The layers are separated and the aqueous phase is extracted with DCM (2x75 mL). The organics are combined and washed with water (2x50 mL). The organic phase is then concentrated under vacuum (crystallization occurs during concentration) to about 20 mL and the rest of the product crystallized out by adding *tert*-butyl methyl ether. The slurry is filtered and dried on high vacuum at room temperature overnight to provide 6.87 g (86%) of **5b** as a tan solid. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ 2.42 (s, 3H), 2.62 (m, 4H), 4.01 (s, 2H), 4.45 (s, 4H), 4.96 (s, 2H), 6.11 (bs, 1H), 7.49 (s, 1H), 7.52 (s, 1H).

Preparation of 7-chloro-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-carbaldehyde, 20a

To a solution of oxalyl chloride (28.8 mL, 330 mmol) and dichloromethane (DCM, 800 mL) at -78 °C is added dimethyl sulfoxide (46.7 mL, 660 mmol) over 6 min. The temperature of the solution increased to -58 °C during the addition and was cooled back to about -70 °C. After stirring for 3 min, the chloro alcohol **5a** (100 g, 275 mmol) was added as a solution in DCM (140 mL) over 10 min. The yellow slurry was stirred for 45 min, then triethylamine (153 mL, 1.10 mol) was added over 4 min. Stirring was continued for 10 min at -78 °C, then the cooling bath was removed. The reaction mixture was allowed to warm to -5 °C and then poured into 2 L of water. The organic layer was separated and the aqueous layer was extracted with DCM (3 x 1.5 L). The combined organic layers were washed with brine (2 L), dried over anhydrous sodium sulfate and concentrated *in vacuo* to give 99.0 g (99%) of product **17a** as a dark solid. This crude product was used for the next step without further purification. Recrystallization from DCM-methanol (5:1) provided an analytical sample as a light yellow solid: mp = 140-142 °C (dec.). ^{1}H NMR ($CDCl_3$, 200 MHz) δ 2.30 (s, 3H), 2.45, 2.59 (m, 8H), 3.98 (s, 2H). 4.41(s, 4H), 7.46 (s, 1H), 7.53 (s, 1H), 10.4 (s, 1H). IR (KBr) 2982, 2797, 1675, 1570, 1500, 1240 cm^{-1}. Elemental analysis: Calculated for $C_{18}H_{20}ClN_3O_3$: C 59.75, H 5.57, N 11.61. Found: C 59.78, H 5.62, N 11.64.

Preparation of 7-iodo-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-carbaldehyde, 17b

To a mixture of chloro aldehyde **17a** (120 g, 346 mmol) and sodium iodide (1.04 kg, 6.92 mmol) in acetonitrile (3 L) was added concentrated hydrogen chloride (59.7 mL, 726 mmol) over 5 min. The white slurry was refluxed for 15 h. The solvent was mostly removed by short path distillation *in vacuo*. The resultant thick slurry was cooled to room temperature and treated with 2.5 L of water and 2.5 L of DCM. The organic layer was separated and the aqueous layer was extracted with DCM (3 x 2 L). The combined organic layers were washed with brine (2.5 L), dried over anhydrous sodium sulfate, concentrated and dried *in vacuo* at rt to afford 110 g (73%) of product **17b** as a yellow solid. This crude product was used for the next step without further purification. Recrystallization from CH_2Cl_2-MeOH (1:1) gave an analytical sample as an off-white solid: mp = 198-200 °C (dec.). ^{1}H NMR ($CDCl_3$, 200 MHz) δ 2.30 (s, 3H), 2.43, 2.57 (m, 8H), 3.93 (s, 2H). 4.40(s, 4H), 7.47 (s, 1H), 7.50 (s, 1H), 10.1 (s, 1H). IR (KBr) 2979, 2798, 1678, 1557, 1500, 1456, 1280, 1105 cm^{-1}. Elemental analysis: Calculated for $C_{18}H_{20}IN_3O_3$: C 47.70, H 4.45, N 9.27. Found: C 47.78, H 4.45, N 9.26.

Preparation of [7-iodo-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-yl]-methanol, 5c

A mixture of iodo aldehyde **17b** (105 g, 232 mmol) as a suspension in methanol (700 mL) was cooled to 0 °C, then sodium borohydride (8.76 g, 236 mmol) was added in three portions over 15 min. The mixture was allowed to warm to room temperature and stirred for 2 h. The solvent was mostly removed *in vacuo* and the

resultant residue was treated with 2.5 L of water and extracted with DCM (4 x 1.5 L). The combined organic layers were washed with brine (2.5 L), dried over anhydrous sodium sulfate and concentrated *in vacuo*. The resultant solid residue was successively treated with 300 mL of DCM and 300 mL of MeOH-EtOAc (1:1). The mixture was swirled at 0 °C for 30 min. The precipitate was collected by filtration, washed with 500 mL of hexane and dried *in vacuo* at rt to afford 87.5 g (83%) of product **5c** as an off-white powder: mp 201-203 °C (dec.). ^{1}H NMR ($CDCl_3$, 200 MHz): δ 2.26 (s, 3H), 2.62 (m, 8H), 4.02 (s, 2H). 4.39(s, 4H), 4.93 (m, 2H), 6.05 (br. s, 1H), 7.49 (s, 1H), 7.51 (s, 1H). HRMS (EI+): Calculated for $C_{18}H_{22}N_3O_3$: 455.0706. Found: 455.0699.

Preparation of [7-bromo-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-yl]-methyl chloride, 18,
and the corresponding quaternary ammonium salt, 19:

A solution ofalcohol **5b** (250 mg, 0.61 mmol) in 6 mL of dichloromethane was cooled to 0 °C and treated with methanesulfonyl chloride (52 uL, 0.67 mmol). After the addition was complete, the mixture was warmed to room temperature and stirred for 4 h. The reaction mixture was partioned between saturated sodium bicarbonate solution (10 mL), the layers were seperated, and the organics were concentrated to yield 238 mg of crude product. This residue was chromatographed (1-4 % methanol/dichloromethane) to provide 107 mg of the chloride **18** as an oil. ^{1}H NMR ($CDCl_3$, 400 MHz): δ 2.27 (s, 3H), 2.57 (m, 8H), 3.96 (s, 2H), 4.39 (s, 4H), 5.15 (s, 2H).

After several days, the sample in the NMR tube precipitated colorless needles which were collected by filtration, dried, and redissolved in methanol-d4. This product was the quaternary ammonium salt, **19**. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ 2.45 (s, 3H), 3.29 (m, 4H), 3.3 (s, 4H), 3.83 (m, 4H), 4.42 (s, 4H), 5.14 (s, 2H), 5.44 (s, 2H), 7.31 (s, 1H), 7.46 (s, 1H).

Preparation of 4(S)-4-Ethyl-4-hydroxy-7-[7-chloro-9-(4-methylpiperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-ylmethyl]-4,7-dihydro-1H-pyrano[3,4-c]pyridine-3,8-dione, 20a:

A mixture of 0.71g of [7-chloro-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-yl]-methanol, 314 mg of 4(S)-4-ethyl-4-hydroxy-4,7-dihydro-1H-pyrano[3,4-c]pyridin-3,8-dione, prepared as described in Intermediate 8, above, and 0.47g of triphenylphosphine is stirred in 2 ml of dichloromethane under nitrogen while 0.28 ml of diethylazodicarboxylate is added dropwise. The mixture is stirred at ambient temperature for 1 hour and the solid is filtered and rinsed with 2 ml of dichloromethane. To the filtrate is added 30 ml of methyl t-butyl ether with stirring. The resulting solid is collected by filtration and rinsed with 4 ml of methyl t-butyl ether to obtained 385 mg of a tan solid. This solid is dissolved in 2 ml of 10% methanol/dichloromethane to which slowly is added 5.5 ml of methyl t-butyl ether with stirring. After 30 minutes, the slurry is cooled to 0 °C for 30 minutes, the solid is collected by filtration and washed with 5 ml of methyl t-butyl ether. After drying, the yield is 281 mg of 4(S)-4-ethyl-4-hydroxy-7-[7-chloro-9-(4-methyl-piperazin-1-yl-methyl)-2,3-dihydro-[1,4]dioxano[2,3-g]quinolin-8-yl]-4,7-dihydro-1H-pyrano [3,4-c]pyridine-

3,8-di-one, **20a** as an off white solid: m.p. (179-183 (dec.)); ^{1}H-NMR ($CDCl_3$, 400 MHz) δ 0.97 (t, J = 7.2 Hz, 3H), 1.81 (m, 2H), 2.17 (s, 3H), 2.43 (m, 8H), 3.82 (s, 2H), 4.41 (m, 4H), 5.44 (ABq, J_{AB} = 16.3 Hz, Δν = 92.1 Hz, 2H), 5.49 (ABq, J_{AB} = 15.3 Hz, Δν = 60 Hz, 2H), 6.42 (d, J = 7.2 Hz, 1H), 7.14 (d, J = 7.2 Hz, 1H), 7.44 (s, 1H), 7.59 (s, 1H). mass spectrum m/z = 555.32, 557 (40% of 555); IR (film) 1800, 1660, 1620, 1570, 1505 cm-1.

Preparation of 4(S)-4-ethyl-4-hydroxy-7-[7-bromo-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-ylmethyl]-4,7-dihydro-1H-pyrano[3,4-c]pyridine-3,8-dione 20b

To a mixture of 4(S)-4-ethyl-4-hydroxy-4,7-dihydro-1H-pyrano[3,4-c]pyridin-3,8-dione **6** (240g, 1.15 moles), [7-bromo-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-yl]-methanol **5b** (609g, 1.49 moles), and triphenylphosphine (361g, 1.38 moles) in dichloromethane (2 L) is added diethylazodicarboxylate (217 ml, 1.38 moles) dropwise with stirring. The reaction warms to reflux during the addition. After the addition is complete, the mixture is stirred for 50 minutes. The mixture is filtered to remove any remaining solids. To the filtrate is added *tert*-butyl methyl ether (3.1 liters) with stirring, causing a precipitate to form. The resultant mixture is cooled to about 1 °C, filtered, and washed with *tert*-butyl methyl ether. The solid is mixed with DCM (2.9 L) and methanol (0.1 L), stirred for 15 minutes and filtered. To the filtrate is added with stirring *tert*-butyl methyl ether (5 L), causing a precipitate to form. The mixture is cooled to 3°C, the solid is collected by filtration, and rinsed with *tert*-butyl methyl ether. After vacuum drying the solid, obtained 492g (71%) of 4(S)-4-ethyl-4-hydroxy-7-[7-bromo-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-ylmethyl]-4,7-dihydro-1H-pyrano[3,4-c]pyridine-3,8-dione, **20b**. MP (183-188 °C (dec.)). ^{1}H-NMR ($CDCl_3$, 400 MHz) δ 0.97 (t, J = 7.2 Hz, 3H), 1.81 (m, 2H), 2.16 (s, 3H), 2.42 (m, 8H), 3.76 (s, 2H), 4.41 (m, 4H), 5.44 (ABq, J_{AB} = 16.3 Hz, Δν = 104.9 Hz, 2H), 5.49 (ABq, J_{AB} = 15.3 Hz, Δν = 81.2 Hz, 2H), 6.42 (d, J = 7.2 Hz, 1H), 7.14 (d, J = 7.2 Hz, 1H), 7.44 (s, 1H), 7.59 (s, 1H). HRMS (EI^+): calc for $C_{28}H_{31}BrN_4O_6$: 589.1427, Found: 589.1441.

Preparation of 4(S)-4-ethyl-4-hydroxy-7-[7-iodo-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-ylmethyl]-4,7-dihydro-1H-pyrano[3,4-c]pyridine-3,8-dione, 23c:

A mixture of pyridone **6** (26.7 g, 128 mmol), iodoalcohol **5c** (58.1g, 128 mmol) and triphenyl phosphine (33.5 g, 128 mmol) in DCM (318 mL) was treated with diethyl azodicarboxylate (20.1 mL, 128 mmol) added dropwise over 15 min. The addition generated heat and gentle reflux of the solvent was observed. The brown solution was allowed to cool to room temperature and stirred for 6.5 h. The solvent was removed in vacuo and the resultant residue was treated with 400 mL of benzene and swirled for 3 min. The so formed precipitate was filtered by suction and washed with 50 mL of cold benzene. The filtrate was concentrated and the resultant solid was chromatographed on silica gel. Elution with 3-50% MeOH in $CHCl_3$ afforded a light yellow solid, which was dissolved in 500 mL MeOH/CH_2Cl_2 (1:100). Recrystallization was then initiated with addition of ethyl acetate. Filtration by suction and drying *in vacuo* afforded 20.0 g of product as a white

powder. The filtrate from the recrystallization was partially concentrated and a second recrystallization gave 18.9 g of product. A third recrystallization in the same fashion yielded 6.1 g of product. Overall, a total of 45.0 g (55%) of product **11** slightly contaminated with ethyl acetate was obtained: mp 158-163 °C (dec.). $[\alpha]_D$ = + 1.9 ° (MeOH, c 1.29). ^{1}H NMR ($CDCl_3$, 200 MHz) δ 0.98 (t, J = 7.3 Hz, 3H), 1.83 (q, J = 7.3 Hz, 2H), 2.19 (s, 3H), 2.15-2.49 (m, 9H), 3.54 (m, 2H), 4.42 (m, 4H), 5.46 (ABq, J_{AB} = 15.4 Hz, Δν = 94.4 Hz, 2H), 5.51 (ABq, J_{AB} = 16.3 Hz, Δν = 110, Hz, 2H), 6.48 (d, J = 7.2 Hz, 1H), 7.20 (d, J = 7.2 Hz, 1H), 7.38 (s, 1H), 7.40 (s, 1H). HRMS (EI+): Calculated for $C_{28}H_{31}IN_4O_6$: 646.1239 Found: 646.1304.

Preparation of 7-(4-Methylpiperazinomethylene)-10,11-ethylenedioxy-20(S)-camptothecin, 4: from chloro quinoline 20a:

A mixture of 100 mg of 4(S)-4-ethyl-4-hydroxy-7-[7-chloro-9-(4-methyl-piper-azin-1-ylmethyl)-2,3-dihydro-[1,4]dioxano[2,3-g]quinolin-8-yl]-4,7-dihydro-1H-pyrano[3,4-c]pyridine-3,8-dione, **20a** 4 mg of palladium(II) acetate, 19 mg of triphenylphosphine, and 37 mg of potassium carbonate in 4 ml of anhydrous acetonitrile was purged with nitrogen and refluxed for 19 hours. At this time the reaction was 99% complete (as monitored by HPLC), was cooled and the resulting solid was collected by filtration, washed with 2 ml of acetonitrile, and resuspended in 2 ml of 10% methanol/dichloromethane. The solid was then removed by filtration, and 4 ml of acetone is added to the filtrate with stirring. After 1 hour, the precipitate which forms was collected by filtration and rinsed with 4 ml of acetone. After drying, the yield was 51 mg (55%) of 7-(4-methylpiperazinomethylene)-10,11-ethylenedioxy-20(S)-camptothecin, **4** free-base.

from bromoquinoline 20b:

After purging with nitrogen, 4(S)-4-ethyl-4-hydroxy-7-[7-bromo-9-(4-methyl-piperazin-1-ylmethyl)-2,3-dihydro-[1,4]dioxino[2,3-g]quinolin-8-ylmethyl]-4,7-dihydro-1H-pyrano[3,4-c]pyridine-3,8-dione **20b** (216 g, 0.36 mol), palladium(II) acetate (4.85 g, 0.0216 mol), powdered potassium carbonate (74.6 g, 0.54 mol), and triphenylphosphine (47.2 g, 0.18 mol) in acetonitrile (9 L) are refluxed under nitrogen for about 12 hours. The mixture is cooled to about 0 °C, filtered, and rinsed with acetonitrile (0.2 L). The crude solid is mixed with DCM (3.25 L) and methanol (550 mL), stirred for about 0.5 hours, filtered, and washed with DCM (0.2 L). The filtrate is treated with triphenylphosphine (54 g) and stirred under nitrogen for about 1.5 hours. Acetone (1.9 L) is added (65 ml/min) with stirring, causing a precipitate to form. The mixture is cooled at 0 °C for 4 hours. The mixture is filtered, and the solid is washed with acetone (0.2 L), and air-dried to give crude product (146 g, 78%). The crude product is dissolved in DCM (2.5 L) and methanol (0.3 L) for 0.5 hours. Triphenylphosphine (36.5 g, 0.39 equiv) is added and the solution is stirred at RT for 2.5 hours under nitrogen. Acetone (1.7 L) is added (85 ml/min) causing a precipitate to form. The resulting mixture is cooled at 0 °C for 4 hours. Filtration, washing with acetone (0.4 L), and drying at 30°C provides cruede product (135 g, 72%). The product is dissolved in DCM (2 L) and methanol (0.31 L). Acetone (1.6 L) is added dropwise to the solution causing a precipitate to form. The slurry is cooled at 0 °C for 4 hours. Filtration, washing with acetone (0.2 L), and

drying at 30 °C provides crude product (124.8 g, 67%). The crude product is dissolved in DCM (1.6 L) and methanol (350 mL). Acetone (1.4 L) is added (280 ml/min) causing a precipitate to form. The slurry is cooled at 0 °C for 3.5 hours. Filtration, washing with acetone (.25 vol/wt), and vacuum-drying at 40 °C provides 115 g (61%) of 7-(4-methylpiperazinomethylene)-10,11-ethylenedioxy-20(S)-camptothecin, **4** free-base as a yellow solid.

from iodo quinoline 20c:

A mixture of iodo quinoline **20c** (30.0 g, 46.4 mmol), palladium(II) acetate (213 mg, 0.928 mmol), anhydrous potassium carbonate powder (12.8 g, 92.8 mmol) and triphenyl phosphine (6.09 g, 23.2 mmol) in 1.8 L of anhydrous acetonitrile was brought to reflux and the iodo olefin was dissolved. As the reflux was continued for 16 h, the product precipitated. After being cooled to 0 °C and stirred for an additional 2.5 h, the mixture was filtered by suction and the resultant yellow cake was treated with 1 L of chloroform . The suspension was filtered and washed with chloform (5 x 200 mL), the combined filtrates were concentrated to 300 mL and treated with 30.0 g of triphenyl phosphine. After being stirred at room temperature for 30 min, the solution was treated with 100 mL of acetone. The resultant precipitate was filtered by suction to yield 17.2 g (72%, >99.5% pure by RP HPLC at 270 nm) of product **4** free-base as a light yellow powder. The product was further purified by stirring as a solution in 220 mL of MeOH/$CHCl_3$ (1:10) containing 5.6 g of triphenyl phosphine, followed by precipitation with 50 mL of acetone. Filtration by suction and drying in vacuo afforded 16.3 g (68%) of product **4** free-base. At this stage, analysis showed 5 ppm of palladium and 17.4 ppm of phosphorus. Further purification was carried out by dissolving the compound in 165 mL of MeOH/$CHCl_3$ (1:10), followed by precipitation with 150 mL of acetone. Filtration and drying *in vacuo* at room temperature gave 15.7 g (65%) of product **4** free-base. Analysis indicated nondetectable amount (< 2ppm) of palladium and phosphorus in the product: mp: 275 °C (dec.). $[\alpha]_D$ = + 22.6 ° ($CHCl_3$, c 1.02). ^1H NMR ($CDCl_3$, 400 MHz): δ 1.04 (t, J = 7.4 Hz, 3H), 1.87 (m, 2H), 2.31 (s, 3H), 2.20-2.59 (m, 9H), 3.97 (s, 2H), 4.46 (s, 4H), 5.32 (s, 2H), 5.55 (ABq, J_{AB} = 16.2 Hz, Δν = 180 Hz, 2H), 7.60 (s, 1H), 7.66 (s, 1H), 7.72 (s, 1H). Elemental analysis: Calculated for $C_{28}H_{30}N_4O_6$: C 64.85, H 5.83, N 10.80. Found: C 64.34, H 5.83, N 10.71.

A sample of material from the filtrates, enriched in impurities was chromatographed with 5% methanol : dichloromethane to give a sample of chloromethylammonium salt **21** (X = OAc^-):

^1H NMR (500 MHz, DMSO-d_6) δ 0.88 (t, J = 7.3 Hz, $CH_2C\underline{H}_3$), 1.78 (s, 3H, $C\underline{H}_3CO_2$-), 1.87 (m, 2H, $CH_3C\underline{H}_2$-), 2.92 (m, 4 H, $CH_2N(C\underline{H}_2CH_2)_2N^+$), 3.20 (s, 3 H, N^+-CH_3), 3.52 (m, 4 H, $CH_2N(CH_2C\underline{H}_2)_2N^+$), 4.14 (dd, J = 13.7 Hz, 2 H, $NC\underline{H}_2Ar$), 4.44 (s, 4 H, $OC\underline{H}_2C\underline{H}_2O$), 5.26 (s, 2H, $CONCH_2$), 5.42 (s, 2 H), 5.54 (br s, 2 H, N^+-CH_2Cl), 7.27 (s, 1 H, CONC=CH), 7.56 (s, 1 H, OC-CH=C-N), 7.77 (s, 1 H, OC-CH=C-C). ^{13}C NMR (123.7 MHz, DMSO-d_6) δ 173.4, 172.5, 156.8, 150.3, 150.1, 147.2, 146.0, 145.1, 136.3, 128.4, 123.5, 118.4, 113.6, 109.4, 96.1, 72.4, 68.4, 65.3, 64.3, 58.1, 53.9, 50.0, 45.1, 30.3, 23.0, 7.7

Preparation of 7-(4-Methylpiperazinomethylene)-10,11-ethylenedioxy-20(S)-camptothecin, 4, GI147211C

A mixture of the free base **4** (14.0 g, 27.0 mmol) in 350 mL of 6 N HCl was brought to reflux for 35 min. A small amount of precipitate was generated after initial complete solution formation. Without being cooled, the mixture was filtered through a Supor (0.45m) filtering memberane. Washing with hot 6N HCl (100 mL) was able to dissolve the above precipitate. The combined filtrates were cooled to 35 °C. Recrystallization was initiated by addition of 20 mL of 200 proof ethanol. After stirring for 1h, 150 mL more ethanol was added. The mixture was allowed to stand at 0 °C for 24 h. Filtration and drying in vacuo at 70 °C yielded 14.5 g (88%) of product as a yellow powder. A ^{1}H NMR shift study with Eu(hfc)$_3$ indicated single enantiomer. mp = 280 °C (dec.). $[\alpha]_D$ = -6.72 ° (H_2O, c 0.67). ^{1}H NMR (D_2O, 400 MHz) δ 0.92(t, J = 7.2 Hz, 3H), 1.89 (m, 2H), 2.55 (m, 2H), 2.81 (s, 3H), 2.96 (m, 2H), 3.11 (m, 2H), 3.41 (m, 3H), 3.71 (s, 2H), 5.32 (ABq, J_{AB} = 16.1 Hz, Δν = 54.2 Hz, 2H), 6.84 (s, 1H), 6.97 (s, 1H), 7.00 (s, 1H). Elemental analysis: Calculated for $C_{28}H_{30}N_4O_6 \cdot 2HCl \cdot H_2O$: C 55.18, H 5.62, N 9.19, Cl 11.63. Found: C 55.41, H 5.70, N 9.24, Cl 11.52.

Acknowledgements: We thank: Dr. Kevin Facchine for NMR spectra; Dr. Andy Steele, Ms. Brenda Shaffei, and Ms. Kimberly Dunn for Pd analysis; Dr. Dan Norwood, Ms. Dixie Fisher, and Dr. Daniel Morgan for mass spectroscopy; and Professor Daniel Comins (North Carolina State University) for helpful discussions.

Notes and References

[1] This paper is dedicated to Professor Samuel J. Danishefsky in recognition of his many contributions to the field of organic chemistry.

[2] Current address: Miles Inc., West Haven, CT 06516-4175

[3] Current address: Trimeris Inc., Durham, NC 27707

[4] Current address: Amgen Inc., Boulder, CO

[5] For a review of clinical data relating to the camptothecins see: W. J. Slichenmyer, E. K. Rowinsky, R. C. Donehower, S. H. Kaufmann, *J. Nat. Cancer Inst.* **1993**, *85*, 271.

[6] Topotecan was approved by the FDA for marketing to treat ovarian cancer on May 29, 1996.

[7] Irinotecan was approved by the FDA for marketing to treat colorectal cancer on June 14, 1996.

[8] W. D. Kingsbury, J. C. Boehm, D. R. Jakas, K. G. Holden, S. M. Hecht, G. Gallagher, M. J. Caranfa, F. L. McCabe, L. F. Faucette, R. K. Johnson, and R. P. Hertzberg, *J. Med. Chem.* **1991**, *34*, 98.

[9] S. Sawada, S. Okajima, R. Aiyama, K. Nokata, T. Furuta, T. Yokokura, E. Sugino, K. Yamaguchi, and T. Miyasaka, *Chem. Pharm. Bull.* **1991**, *39*, 1446.

[10] M.J. Luzzio, J.M. Besterman, D.L. Emerson, M.G. Evans, K. Lackey, P.L. Leitner, G. McIntyre, B. Morton, P.L. Myers, M. Peel, J. M. Sisco, D.D. Sternbach, W.-Q. Tong, A. Truesdale, D.E. Uehling, A. Vuong, and J. Yates, *J. Med. Chem.* **1995**, *38*, 395.

[11] M. C. Wani, A. W. Nicholas, M. E. Wall, *J. Med. Chem.* **1987**, *30*, 2317.

[12] (a) D. P. Curran et al, *Angew. Chem. Int. Ed.* Engl. **1995**, *34*, 2683. (b) D. P. Curran, H. Liu, H. Josien, and S.-Bo Ko, *Tetrahedron*, **1996**, *52*, 11385.

[13] a) F. G. Fang, M. W. Lowery, and S. Xie, U.S. Patent 5,491,237 (2/13/96). b) F. G. Fang, S. Xie, and M. W. Lowery, *J. Org. Chem.* **1994**, *59*, 6142.

[14] For reviews on synthetic efforts in this field see: (a) D. P. Curran, J. Sisko, and P. E. Yeske, *Pure Appl. Chem.* **1993**, *65*, 1153. (b) J. C. Cia, C. R. Hutchinson, in *The Alkaloids: Chemistry and Pharmacology*, A. Brossi, Ed., Academic Press: New York, **1983**, vol. 21, p 101. (d) C. R. Hutchinson, *Tetrahedron* **1981**, 37, 1047. (e) A. G. Schultz, *Chem. Rev.* **1973**, *73*, 385.

15 For recent synthetic studies towards **1** see: (a) M. Ciufolini and F. Roschangar, *Angew. Chem. Int. Ed. Engl.* **1996**, *35*, 1692. (b) J. M. D. Fortunak, J. Kitteringham, A. R. Mastrocola, M. Mellinger, N. J. Sisti, J. L. Wood, and Z.-P. Zhuang, *Tetrahedron Lett.* **1996**, *37*, 5683. (c) J. M. D. Fortunak, A. R. Mastrocola, M. Mellinger, N. J. Sisti, J. L. Wood, and Z.-P. Zhang, *Tetrahedron Lett.* **1996**, *37*, 5679. (d) S.-s. Jew, K.-d. Ok, H.-j. Kim, M. G. Kim, J. M. Kim, J. M. Hah, Y.-s. Cho, *Tetrahedron: Asymmetry* **1995**, *6*, 1245. (e) D. H. Comins, H. Hong, J. K. Saha, and G. Jianhua, *J. Org. Chem.* **1994**, *59*, 5120. (f) W. Shen, C. A. Coburn, W. G. Bornman, and S. J. Danishefsky, *J. Org. Chem.* **1993**, *58*, 611.

16 D.L. Comins, M.F. Baefsky, and H. Hong, *J. Am. Chem. Soc.* **1992**, *114*, 10971.

17 For a recent review see: R. Grigg, V. Sridharan, P. Stevenson, S. Sukirthalingam, and T. Worakun, *Tetrahedron* **1990**, *46*, 4003.

18 For a recent review see: H. C. Kolb, M. S. VanNieuwenhze, and K. B. Sharpless, *Chem. Rev.* **1994**, *94*, 2483.

19 For some recent results on the intramoleular Heck reaction (cf. **9** to **10**) see: D. D. Bankston, F. G. Fang, and S. Xie, *J. Org. Chem.*, manuscript in preparation.

20 I. N. Houpis, A. Molina, A. W. Douglas, L. Xavier, J. Lynch, R. P. Volante, and P. J. Reider, *Tetrahedron Lett.* **1994**, 35, 6811.

21 T. Sugasawa, M. Adachi, K. Sasakura, A. Kitagawa, *J. Org. Chem.* **1979**, *44*, 578.

22 cf. J. A. Robl, *Synthesis*, **1991**, 56.

23 For reviews see: (a) O. Mitsunobu, *Synthesis* **1981**, 1. (b) D. L. Hughes, *Org. React.* **1992**, *42*, 335.

24 At the time we implemented this solution, no reports on the Mitsunobu reaction of pyridones had appeared in the literature. Subsequently, Comins has published a study on the Mitsonubu reaction of pyridones and the application of this reaction to the synthesis of **1**: (a) D. L. Comins and G. Jianhua, *Tetrahedron Lett.* **1994**, *35*, 2819. (b) D. L. Comins, H. Hong, and G. Jianhua, *Tetrahedron Lett.* **1994**, *35*, 5331. See also reference 15e.

25 For a review see: R. F. Heck, *Org. React.* **1982**, *27*, 345.

26 For a nickel-mediated cyclization of a chloroaromatic system see: M. Mori, S. Kudo, and Y. Ban, *J. Chem. Soc., Perkin Trans. 1* **1979**, 771.

27 A similar strategy for palladium removal has been developed in the synthesis of the angiotensin II receptor antagonist losartan: R. D. Larsen, A. O. King, C. Y. Chen, E. G. Gorley, B. S. Foster, F. E. Roberts, C. Yang, D. R. Lieberman, R. A. Reamer, D. M. Tschaen, T. R. Verhoeven, P. J. Reider, Y. S. Lo, L. T. Rossano, A. S. Brookes, D. Meloni, J. R. Moore, and J. F. Arnett, *J. Org. Chem.* **1994**, *59*, 6391.

28 The biological activity of this compound will be reported elsewhere.

29 N. Menschutkin, *Z. Phys. Chem.* **1890**, *6*, 41.

(*Received* 27 *September* 1996; *accepted* 10 *January* 1997)