

An Synthesis of (Z)-4-Hydroxy-N-Desmethyldesmethyltamoxifen (Endoxifen)

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Supporting Information

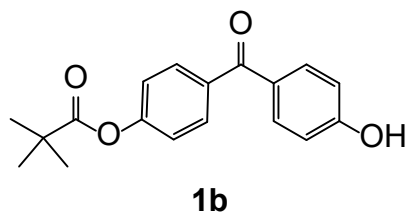
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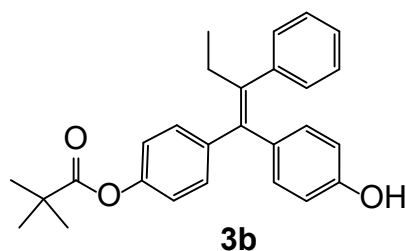
NMR and MS spectra for the intermediates leading to endoxifen, and its (*E*)-Isomer.....S15-S30

Part 1

Experimental Section:

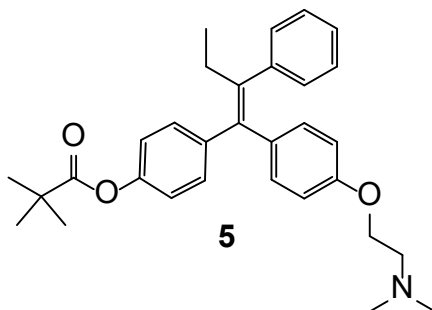


This compound was synthesized following a literature procedure.⁸



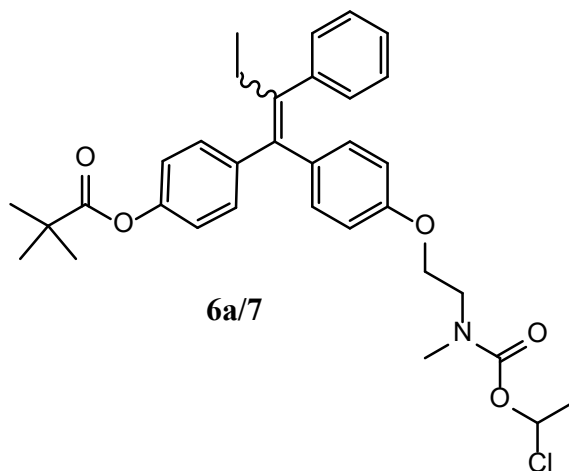
(E)-4-(1-(4-hydroxyphenyl)-2-phenylbut-1-en-1-yl)phenyl pivalate (3b): This compound was synthesized following a literature procedure.⁸

4-(4-hydroxybenzoyl)phenyl pivalate (1b): This compound was synthesized following a literature procedure.⁸



(E)-4-(1-(4-(2-(dimethylamino)ethoxy)phenyl)-2-phenylbut-1-en-1-yl)phenyl pivalate (5)

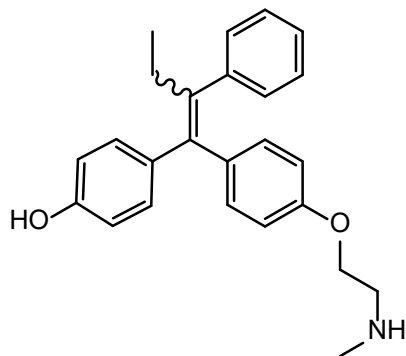
This compound was made by a reported procedure⁸.



(E) and (Z)-4-(1-(4-(2-(((1-chloroethoxy)carbonyl)(methyl)amino)ethoxy)phenyl)-2-phenylbut-1-

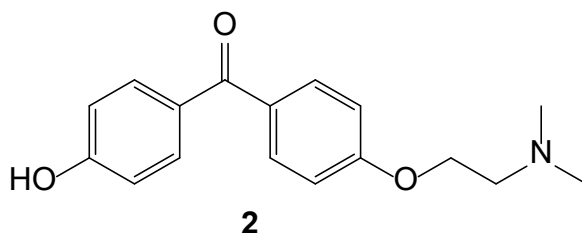
enyl)phenyl pivalate (6b) and (7): 1-chloroethyl chloroformate (0.732 mL, 6.78 mmol) was added dropwise to a solution of (E)-4-(1-(4-(2-(dimethylamino)ethoxy)phenyl)-2-phenylbut-1-enyl)phenyl pivalate **5a** (0.533 g, 1.13 mmol) in dichloroethane (15 mL) at 0° C. The solution was stirred for 10 min at 0° C and then heated to reflux for 12 hrs. The reaction mixture was cooled to rt and solvent was removed under reduced pressure yielding a pale yellow crude oil which was subjected to silica-gel column chromatography using 25% EtOAc/hexane. Fractions containing the desired compounds were evaporated to afford a white solid (0.505g, 79%) containing the desired (E)- and the undesired (Z)-isomer in a 4:1 ratio. Most ¹H NMR signals of the minor isomer were mixed with those of the major isomer except a few that were used for determining isomeric composition. The extracted ¹H NMR of the major isomer: ¹H NMR (300 MHz, CDCl₃): δ 7.18-6.96 (m, 9H), 6.78 (d, 2H, *J* = 8.4 Hz), 6.50 (m, 1H) 6.45 (d, 2H, *J* = 8.4 Hz), 3.91 (m, 2H), 3.48 (m, 2H), 2.94 (d, 3H, *J* = 2.4 Hz), 2.38 (q, 2H,

$J = 7.4$ Hz), 1.70 (m, 3H), 1.29 (s, 9H), 0.83 (t, $J = 7.4$ Hz). MS m/z (ESI) 364.30 (M+1)⁺. HRMS calc'd for C₃₃H₃₉ClNO₅ 564.2517, found 564.2508.



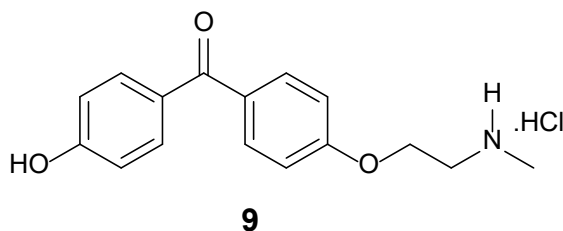
8a (enodxifen)/8b (*E*)-isomer

Endoxifen ((*Z*)-isomer, 8a)--through pivaloyl protection: MeLi (6.7 mL of a solution 1.6 M in ether, 10.74 mmol) was added to a solution of the 4:1 *E/Z* mixture of 4-(1-(4-(2-(((1-chloroethoxy)carbonyl)(methylamino)-ethoxy)phenyl)-2-phenylbut-1-enyl)phenyl pivalate (0.505g, 0.895 mmol) in dry THF (30 mL) at -78 °C. The reaction mixture was stirred for 2 h, quenched with saturated ammonium chloride (2 mL), allowed to warm to rt, and extracted with ethyl acetate (4 × 25 mL). The combined organic phase was dried and concentrated. The crude was purified over silica-gel column chromatography using 15% MeOH/DCM gave endoxifen (0.271 g, 81%) with a 4:1 *Z/E* ratio as a yellowish solid from which pure endoxifen was isolated by RP-HPLC as described for endoxifen **6b** and its (*E*)-isomer **7b** (vide infra).



4-(2-(Dimethylamino)ethoxy)phenyl(4-hydroxyphenyl)methanone (2):

Compound **2** was synthesized by following a literature procedure in 46% yield (lit. 40%).⁸ ¹H NMR (300 MHz, CDCl₃): δ 7.76 (d, 2H, *J* = 8.4 Hz), 7.69 (d, 2H, *J* = 8.4 Hz), 6.97 (d, 2H, *J* = 8.0 Hz), 6.88 (d, 2H, *J* = 8.0 Hz), 4.16 (t, 2H, *J* = 5.2 Hz), 2.82 (t, 2H, *J* = 5.2 Hz), 2.37 (s, 6H). MS *m/z* (ESI) 286.34 (*M*+1)⁺. ¹³C NMR (CDCl₃) δ 45.8, 58.9, 66.8, 115.2, 116.2, 130.3, 132.2, 133.3, 133.7, 163.6, 196.8.

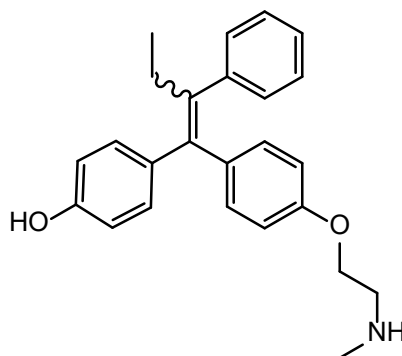


(4-Hydroxyphenyl)(4-(2-(methylamino)ethoxy)phenyl)methanone

hydrochloride (9): 1-chloroethyl chloroformate (1.14 mL, 10.51 mmol) was added dropwise to a solution of 4-(2-(dimethylamino)ethoxy)phenyl(4-hydroxyphenyl)methanone **2** in dichloroethane (13.8 mL) at 0° C. The resulting solution was stirred for 10 min at 0 °C and heated to reflux for 12 h. Cooling the reaction mixture to room temperature and removing the solvent under reduced

pressure give a yellowish crude oil. This residue was dissolved in MeOH (20 mL) and 6M HCl (5 mL) and stirred under reflux for 12 h and then cooled to room temperature. The solvent was removed under reduces pressure, sat'd NaHCO₃ solution was added and the residue was extracted with EtOAc (3 x 25 mL). The combined organic extracts were washed with sat'd NaHCO₃, water, and brine, dried over MgSO₄, filtered and concentrated to afford crude **9** which was purified by silica-gel column chromatography using 0.1% TEA in 10% MeOH/DCM (0.394 g, 83% yield). The free amine was converted to a hydrochloride by treating it with 6N HCl-dioxane.

¹H NMR (300 MHz, MeOD₄): δ 7.73 (d, 2H, *J* = 8.4 Hz), 7.66 (d, 2H, *J* = 8.4 Hz), 7.07 (d, 2H, *J* = 8.3 Hz), 6.88 (d, 2H, *J* = 8.3 Hz), 4.20 (t, 2H, *J* = 4.9 Hz), 3.04 (t, 2H, *J* = 4.9 Hz), 2.50 (s, 3H). MS *m/z* (ESI) 272.15 (M+1)⁺. ¹³C NMR (75 MHz, CD₃OD): δ 33.9, 64.6, 115.3, 116.2, 130.2, 133.1, 133.2, 133.7, 162.5, 163.5, 196.7. HRMS calculated for C₁₆H₁₈NO₃ 272.1287, found 272.1273.



8a (enodxifen)/8b (*E*)-isomer

Synthesis of (Z)- (endoxifen, 8a) and (E)-isomer (8b)—without pivalate protection: To a suspension of zinc dust (1.02 g, 15.60 mmol) in anhydrous THF (15 mL) was added TiCl_4 (0.855 mL, 7.80 mmol) dropwise under argon at room temperature and the mixture refluxed for 2 h. A suspension of (4-hydroxyphenyl)(4-(2-(methylamino)ethoxy)phenyl)methanone.HCl (0.60 g, 1.949 mmol) and propiophenone (0.907 mL, 6.82 mmol) in anhydrous THF (10 mL) was added at once and the reflux was continued for 5 h. Upon completion, the reaction mixture was cooled to room temperature, quenched with 10% K_2CO_3 (~ pH 8) and extracted with EtOAc (3 x 25 mL). The combined organic extracts were successively washed with 10% K_2CO_3 (2 x 15 mL) and brine, dried over MgSO_4 , filtered, and the filtrate concentrated under reduced pressure to afford a crude that was purified by 10% MeOH/DCM to afford a mixture of endoxifen and its (*E*)-isomer (0.655 g, 90% combined yield). Endoxifen and its (*E*)-isomer were separated by reverse phase HPLC (Vydac column, C-8, 2.2 x 25 cm, elution with 30% aqueous 20 mM triethylammonium bicarbonate buffer in acetonitrile (pH 8.8) in isocratic elution mode; FR 8 mL/min, λ_{max} = 254 nm. RT = 53 min for endoxifen and 81 min for (*E*) isomer). Smaller quantities can also be separated easily by using overloads on a smaller columns (PrincetonSPHER-300, C8, 5m, column, C-8, 15x1 cm, elution with 60% aqueous 20 mM triethylammonium bicarbonate buffer in acetonitrile (pH 8.8) in isocratic elution mode; FR 2 mL/min, λ_{max} = 254 nm. RT = 42 min for endoxifen and 60 min for (*E*) isomer). The combined HPLC fractions were evaporated and treated with saturated NaHCO_3 solution and extracted with DCM (2 x 25 mL). The DCM extracts were combined,

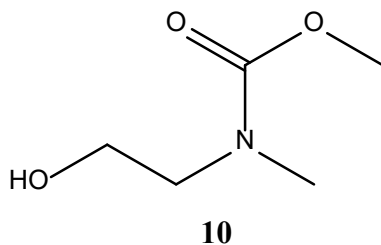
washed with water, dried over MgSO_4 , and concentrated under reduced pressure to furnish endoxifen and its (-)-isomer in pure form as solids. Endoxifen (**8a**): ^1H NMR (300 MHz, CDCl_3): δ 7.17-7.07 (m, 5H), 6.99 (d, 2H, $J = 8.4$ Hz), 6.75-6.70 (m, 4H), 6.48 (d, 2H, $J = 8.7$ Hz), 3.95 (t, 2H, $J = 4.8$ Hz), 2.93 (t, 2H, $J = 4.8$ Hz), 2.49-2.34 (m, 5H), 0.90 (t, $J = 7.3$ Hz). MS m/z (ESI) 374.30 ($\text{M}+1$) $^+$. ^1H NMR ($\text{DMSO}-d_6$): δ 7.06-7.18 (m, 5H), 6.98 (d, 2H, $J = 8.3$ Hz), 6.96-6.76 (m, 4H), 6.56 (d, 2H, $J = 8.5$ Hz), 3.85 (t, 2H, $J = 5.4$ Hz), 2.72 (t, 2H, $J = 5.4$ Hz), 2.43 (q, 2H $J = 7.3$ Hz), 2.27(s, 3H), 0.84 (t, 3H $J = 7.3$ Hz). MS m/z (ESI) 374.30 ($\text{M}+1$) $^+$. ^{13}C NMR ($\text{DMSO}.d_6$) δ 13.4, 28.50 36.0, 50.1, 66.8, 113.3, 114.9, 125.9, 127.8, 129.3, 130.0, 131.4, 133.9, 135.4, 137.9, 139.9, 142.2, 156.0, 156.4; HRMS calculated for $\text{C}_{25}\text{H}_{28}\text{NO}_2$ 374.2120, found 374.2105.

(*E*)-isomer of endoxifen (**8b**): ^1H NMR (300 MHz, CDCl_3): δ 7.19-7.08 (m, 5H), 6.82 (d, 2H, $J = 8.5$ Hz), 6.72-6.65 (m, 4H), 6.43 (d, 2H, $J = 8.5$ Hz), 4.07 (t, 2H, $J = 4.9$ Hz), 2.99 (t, 2H, $J = 4.9$ Hz), 2.52-2.45 (m, 5H), 0.91 (t, 3H, $J = 7.2$ Hz). MS m/z (ESI) 374.31 ($\text{M}+1$) $^+$. ^1H NMR ($\text{DMSO}-d_6$): δ 7.13-7.25 (m, 7H), 7.00 (d, 2H, $J = 8.6$ Hz), 6.65 (d, 2H, $J = 8.5$ Hz), 6.46 (d, 2H, $J = 8.5$ Hz), 4.17 (t, 2H, $J = 5.2$ Hz), 2.99 (t, 2H, $J = 5.2$ Hz), 2.51(s, 3H), 2.43 (q, 2H $J = 7.2$ Hz), 0.91 (t, 3H $J = 7.2$ Hz). MS m/z (ESI) 374.31 ($\text{M}+1$) $^+$. ^{13}C NMR ($\text{DMSO}.d_6$) δ 13.4, 28.4, 34.7, 49.0, 65.4, 114.2, 114.3, 125.9, 127.8, 129.3, 130.1, 131.3, 133.56, 136.0, 137.8, 139.8, 142.2, 155.2, 156.9; $\text{C}_{25}\text{H}_{28}\text{NO}_2$ 374.2120, found 374.2114.

Conversion of *E*-isomer (8b) to a 1:1 mixture of endoxifen (8a) and (*E*)-Isomer (8b): The combined (*E*)-isomer fractions from HPLC runs were allowed to equilibrate with 6 N HCl (1:1) at 60 °C for 6 h. resulted in clean conversion of

E-isomer into a 1:1 mixture of *Z*- and *E*-isomers as determined by HPLC chromatogram and ¹H NMR analysis. The solvent was removed under reduced pressure and the pH of the mixture was adjusted to ~ 7.2. Extraction of the mixture with EtOAc (3 x), washing of the combined extracts with water and brine, drying (MgSO₄), and evaporation of the volatiles gave a clean colorless oil that was immediately subjected to semipreparative RP-HPLC separation as described above resulting in enhanced overall yield of endoxifen.

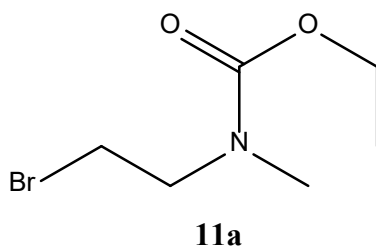
Alternatively, the (*E*)-isomer **8b** was dissolved in a 1:1 mixture of trifluoroacetic acid and dichloromethane and stirred for one hour at ambient temperature. The volatiles were evaporated and the residue was subjected to the HPLC separation as described above.



Ethyl 2-hydroxyethyl(methyl)carbamate (10):

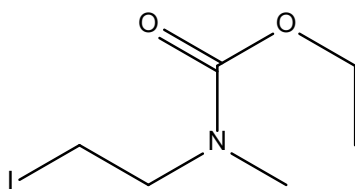
Ethyl chloroformate (9.45 mL, 121.00 mmol) was added dropwise to a solution of 2-(methylanino)ethanol (10.65 mL, 133.00 mmol) and triethylamine (16.87 mL, 121.00 mmol) in DCM (154 mL) at 0° C. The resultant reaction mixture was stirred for 10 min at 0° C and then warmed to room temperature during 2 h. After quenching the reaction mixture with 1M HCl (20 mL), the reaction mixture was extracted with DCM (2 x 25 mL), and the combined extracts were washed with

water, brine, dried over MgSO_4 . Filtration and concentration of the solvent afforded a clear oil (70%, 12.47 g). ^1H NMR (300 MHz, CDCl_3): δ 4.16 (q, 2H, $J = 7.0$ Hz), 3.74 (brs, 2H), 3.44 (brs, 2H), 2.96 (s, 3H), 1.26 (t, 3H, $J = 7.0$ Hz). m/z (ESI) 148.25 ($\text{M}+1$) $^+$. ^{13}C NMR (75 MHz, CDCl_3): δ 14.6, 4.10, 51.6, 61.4, 67.9, 157.7; HRMS calc'd for $\text{C}_6\text{H}_{14}\text{NO}_3$ 148.0974, found 148.0963.



Ethyl 2-bromoethyl(methyl)carbamate (11a):

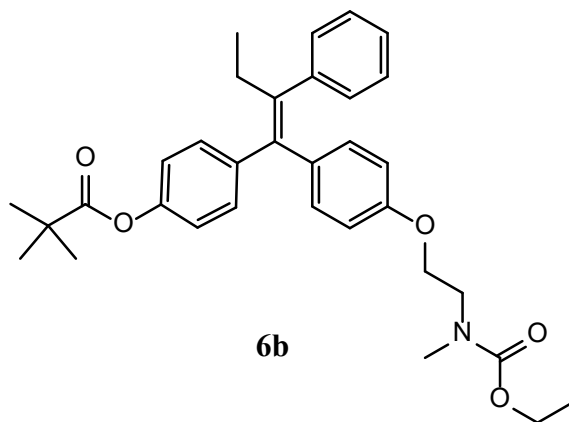
To a solution of ethyl 2-hydroxyethyl(methyl)carbamate **10** (1.00 g, 6.79 mmol) and carbon tetrabromide (4.51 g, 13.59 mmol) in ether (56.40 mL) was added triphenylphosphine (3.56 g, 13.59 mmol) under nitrogen atmosphere and the resulting mixture was stirred for 6 h at room temperature. Hexanes (400 mL) were then added and the reaction mixture was filtered through Celite. The filtrate was evaporated and the residue was directly purified by flash chromatography using 15% ethyl acetate/hexane as eluent to afford **11a** as clear oil (91%, 1.29 g). ^1H NMR (300 MHz, CDCl_3): δ 4.15 (q, 2H, $J = 7.0$ Hz), 3.62 (brs, 2H), 3.48-3.43 (brm, 2H), 2.98 (s, 3H), 1.26 (t, 3H, $J = 7.0$ Hz). m/z (ESI) 210.13 ($\text{M}+1$) $^+$. ^{13}C NMR (75 MHz, CDCl_3): δ 14.6, 29.2, 35.3, 50.4, 61.5, 156.4; HRMS calc'd for $\text{C}_6\text{H}_{13}\text{BrNO}_2$ 210.0130, found 210.0115.



11b

Ethyl 2-iodoethyl(methyl)carbamate (11b):

To a solution of 2-hydroxyethyl(methyl)carbamate **10** (2.5 g, 16.99 mmol) and imidazole (5.78 g, 85.00 mmol) in THF (364 mL) precooled to 0 °C (ice-bath) were added sequentially triphenylphosphine (6.68 g, 25.5 mmol) and iodine (6.47, 25.5 mmol) under nitrogen atmosphere. After 1h of stirring at room temperature, ethyl acetate and saturated aqueous NaHCO₃ were added and the organic layer was separated. Washing the organic layer with saturated Na₂S₂O₃, drying (MgSO₄) the volatiles were evaporated under reduced pressure to a pale yellow residue which was purified by flash chromatography using 15% ethyl acetate/hexane as eluent to give a colorless oil (89%, 3.9 g). ¹H NMR (300 MHz, CDCl₃): δ 4.15 (brq, 2H, *J* = 6.5 Hz), 3.62 (brd, 2H, *J* = 5.7 Hz), 3.25-3.18 (brm, 2H), 2.95 (s, 3H), 1.27 (t, 3H, *J* = 7.0 Hz). *m/z* (ESI) 257.87 (M+1)⁺, 289.93 (M+Na)⁺. ¹³C NMR (75 MHz, CD₃OD): δ 13.4, 33.6, 50.0, 50.5, 60.2, 154.7; HRMS calc'd for C₆H₁₃INO₂ 257.9991, found 257.9976.



(E)-4-(1-(4-(2-(ethoxycarbonyl(methyl)amino)ethoxy)phenyl)-2-phenylbut-1-enyl)phenyl pivalate (6b**)**

This compound was synthesized in three ways.

Method A: To a solution of 2-hydroxyethyl(methyl)carbamate **10** (0.046 g, 0.312 mmol) and (*E*)-4-(1-(4-hydroxyphenyl)-2-phenylbut-1-enyl)phenyl pivalate **3b** (0.100 g, 0.25 mmol) in THF (1 mL) was added triphenylphosphine (0.164 g, 0.624 mmol) and the mixture was cooled to 0 °C and diisopropyl azodicarboxylate (0.098 mL, 0.499 mmol) was added to it dropwise. The solution was stirred for 3 h at room temperature. The solvent was removed under reduced pressure and the residue was directly purified by flash chromatography (eluent: 10-15% ethyl acetate/hexane) to afford the pure isomer **6b** as a clear oil (41%, 0.054 g). ¹H NMR (300 MHz, CDCl₃): δ 7.24-7.03 (m, 9H), 6.75 (d, 2H, J = 8.7 Hz), 6.51 (d, 2H, J = 8.7 Hz), 4.10 (q, 2H, J = 7.1 Hz), 3.96 (brs, 2H), 3.55 (brs, 2H), 2.97 (s, 3H), 2.46 (q, 2H, J = 7.3 Hz), 1.36 (s, 9H), 1.27 (brt, 3H, J = 7.1 Hz), 1.27 (t, 3H, J = 7.1 Hz). m/z (ESI) 530.27 (M+1)⁺. ¹³C NMR (75 MHz, CDCl₃): δ 13.5, 14.71, 27.1, 29.0, 35.9, 39.1, 61.3, 113.3, 114.0, 120.3, 121.1, 126.1, 127.9, 129.7

130.4, 130.7, 131.7, 132.0, 137.3, 1414.1, 142.3, 149.7, 177.1; HRMS calc'd for $C_{33}H_{40}NO_5$ 530.2906, found 530.2893.

Method B: To a solution of (*E*)-4-(1-(4-hydroxyphenyl)-2-phenylbut-1-enyl)phenyl pivalate **3b** (0.100 g, 0.25 mmol) in DMF (1 mL) was added Cs_2CO_3 (0.122 g, 0.375 mmol) and ethyl 2-iodoethyl(methyl)carbamate **11b** (0.096 g, 0.375 mmol) at room temperature under argon atmosphere. The resulting mixture was stirred at the same temperature for 16 h. Upon completion, the reaction mixture was quenched with saturated aqueous NH_4Cl and extracted with diethyl ether (3 x 5 mL). The combined organic extracts were washed with brine, dried over $MgSO_4$, filtered and concentrated. Flash chromatography of the residue using 10-15% ethyl acetate/hexane as eluent furnished a 8:2 mixture of **6b** and **7a** (as determined by comparison with the pure product **6b** obtained above by Mitsunobu reaction) as a clear oil (45%, 0.06 g).

Method C: To a solution of (*E*)-4-(1-(4-hydroxyphenyl)-2-phenylbut-1-enyl)phenyl pivalate **3b** (0.100 g, 0.25 mmol) in DMF (1 mL) was added Cs_2CO_3 (0.122 g, 0.375 mmol) and the resulting suspension was heated at 50 °C. To this was added ethyl 2-bromoethyl(methyl)carbamate (0.079 g, 0.375 mmol) and stirred at same temperature under argon atmosphere for 16 h. Upon completion the reaction mixture was quenched with saturated aqueous NH_4Cl and extracted with diethyl ether (3 x 5 mL), the combined organic extracts were washed with brine, dried over $MgSO_4$, filtered and concentrated. Elution of the product 10-15% ethyl acetate/hexane over silica gel gave a 8:2 mixture of **6b** and **7b** Flash

chromatography of the residue using 10-15% ethyl acetate/hexane as eluent furnished a 8:2 mixture of **6b** and **7b** as a clear oil as a clear oil (38%, 0.05 g).