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Antibody-Catalyzed Enantioselective Norrish Type II Cyclization

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Supporting Information

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Experimental Information

General methods.

^{1}H NMR spectra were recorded on a Bruker AM 200 spectrometer, operating at 200 MHz using $CDCl_3$ as a solvent (unless otherwise specified). EI-MS spectra were measured on a Finnigan MAT-711 spectrometer. CI-MS spectra were measured on a Finnigan TSQ-70 GCMS-CI linked to a Varian GC equipped with a DB-5ms capillary column. UV-vis spectra were recorded on a Shimadzu UV-1601 spectrometer. Quartz Ultra micro UV Spectrophotometer cuvets from Sigma-aldrich C-9792 were used for measuring antibodies concentrations. Irradiation experiments performed with cuvets were carried out in Fischerbrand 0.100 cm, SCC 282 cuvets. TLC was performed on glass sheets pre-coated with silica gel (Merck, Kieselgel 60, F254, Art. 5715). Column chromatographic separations were performed on silica gel (Merck, Kieselgel 60, 230-400 mesh, Art. 9385) under pressure. THF was dried and distilled over sodium/benzophenone. HPLC analyses were carried out with a Merck-Hitachi Lachrom system equipped with a L-7100 pump, L-7400 UV-vis detector and D-7000 system manager using Supelco RP LC-18 analytical column.

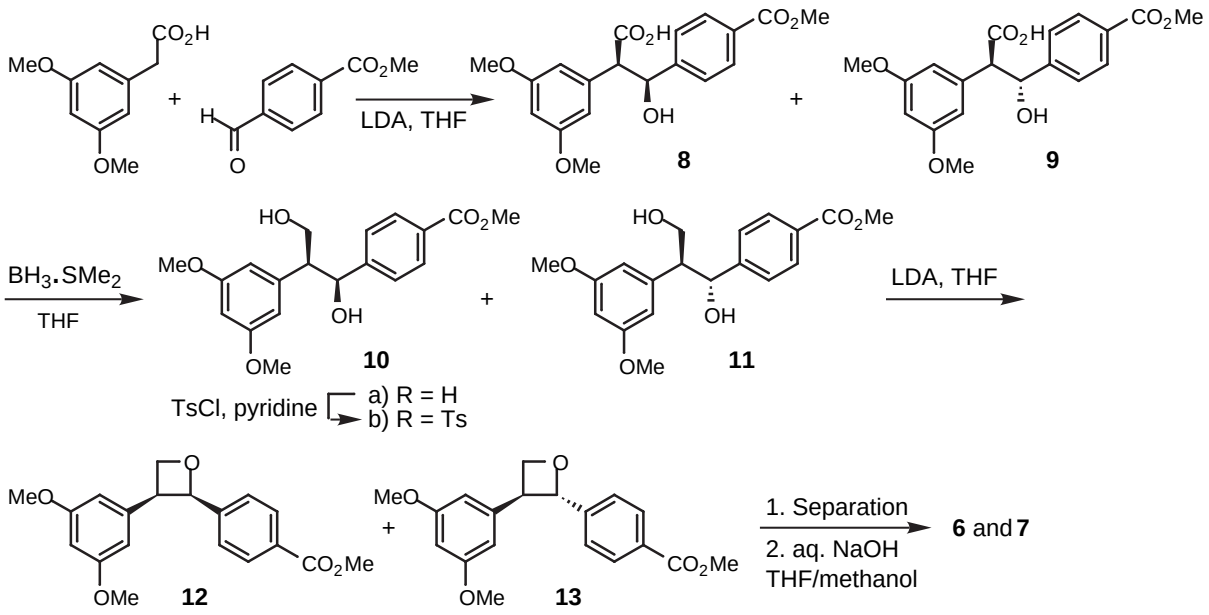

Figure 1: Synthesis of haptens 6 and 7.

Methyl 4-[2-carboxy-2-(3,5-dimethoxy-phenyl)-1-hydroxy-ethyl]-benzoate, 8, 9. A Solution of LDA was prepared by dropwise addition of *n*-BuLi (2.5 M, 8.8 mL, 22 mmol) to a solution of diisopropylamine (4.7 g, 24 mmol) in dry THF (30 mL) at 0 ˚C. A Solution of the 3,5-dimethoxy phenyl acetic acid (1.96 g, 10 mmol) in dry THF (5 mL) was added dropwise to the LDA solution at –78 ˚C and stirring was continued for 2 h while warming from –78 ˚C to 0 ˚C. A Solution of methyl 4-formyl-benzoate (1.64 g, 10 mmol) in dry THF (5 mL) was added to the reaction mixture at –78 ˚C. After 0.5 h, the reaction was worked up by addition of saturated aq. NH_4Cl followed by 3N HCl and extraction with ethyl acetate. The organic layer was washed with brine, dried over Na_2SO_4 and the solvent was removed under reduced pressure to give a mixture of **8** and **9**, which was taken to next step without purification. ^{1}H NMR ($CDCl_3$, 200 MHz): δ 7.82-7.60 (m, 2H), 7.20-6.85 (m, 2H), 6.40-6.05 (m, 3H), 5.35-5.05 (m, 1H), 3.85 and 3.83 (s each, together 3H), 3.48 and 3.41 (s each, together 6H), 3.65-3.50 (m, 1H).

Methyl 4-[2-(3,5-dimethoxy-phenyl)-1,3-dihydroxy-propyl]-benzoate, 10, 11. A Solution of $BH_3.SMe_2$ (2M in THF, 10 mL) was added dropwise to a solution of the above-mentioned mixture of **8** and **9** and the reaction mixture was stirred overnight at room temperature. Solvents were removed under reduced pressure and the residue was purified by column chromatography (silica gel, hexane-ethyl acetate) to afford a mixture of diols **10** and **11** (2.3 g, 66% for both steps). ^{1}H NMR ($CDCl_3$, 200 MHz): δ 7.91 and 7.80 (d, J = 8.2 Hz each, together 2H), 7.27 and 7.15 (d, J = 8.2 Hz each, together 2H), 6.35-6.05 (m, 3H), 5.04 and 4.96 (d each, J = 6.2 and 7.2 Hz, together 1H), 3.86 and 3.83 (s each, together 3H), 3.73 and 3.68 (s each, together 3H), 3.70 and 3.62 (s each, together 3H), 3.95-3.60 (m, 2H), 3.05 (m, 1H), 2.65 (br s, 2H).

Methy 4-[3-(3,5-Dimethoxy-phenyl)-oxetan-2-yl]-benzoate, 12, 13. TsCl (1.01 g, 5.3 mmol) was added to a solution of the mixture of **10** and **11** (1.53 g, 4.42 mmol) in dry pyridine (15 mL) at 0 ˚C. After stirring for 16 h, the mixture was worked up with water and CH_2Cl_2. The organic layer was washed with dilute HCl and with brine. Solvents were removed under reduced pressure and the residue was filtered through silica gel (hexane-ethyl acetate) to afford a mixture of tosylates **10b** and **11b** (1.83 g, 83%).

A freshly prepared LDA solution in THF (4 mmol) was added dropwise to a solution of the mixture of **10b** and **11b** (1.83 g) in dry THF (20 mL) at 0 ˚C. The mixture was stirred at 60 ˚C for 0.5 h, then cooled to room temperature and worked up with saturated aq. NH_4Cl and ether. The combined organic layer was washed with brine, dried over Na_2SO_4 and solvent was removed under reduced pressure. The crude residue was separated over silica gel (hexane-ethyl acetate) to afford compounds **12** and **13** separately. ^{1}H NMR of **12** ($CDCl_3$, 200 MHz): δ 7.85 (d, J = 8.0 Hz, 2H), 7.17 (d, J = 8.0 Hz, 2H), 6.20 (d, J = 8.6 Hz, 1H), 6.15 (d, J = 2.0 Hz, 2H), 6.14 (t, J = 2.0 Hz, 1H), 5.14 (dd, J = 8.6, 6.5 Hz, 1H), 4.84 (t, J = 6.6 Hz, 1H), 4.51 (q, J = 6.6 Hz, 1H), 3.84 (s, 3H), 3.58 (s, 6H). ^{1}H NMR of **13** ($CDCl_3$, 200 MHz): δ 8.05 (d, J = 8.3 Hz, 2H), 7.47 (d, J = 8.3 Hz, 2H), 6.52 (d, J = 2.0 Hz, 2H), 6.38 (t, J = 2.0 Hz, 1H), 5.78 (d, J = 6.9 Hz, 1H), 4.91 (m, 2H), 3.93 (m, 1H), 3.90 (s, 3H), 3.79 (s, 6H).

Structure determination of oxetanes 12 and 13. In Analogy to the reports on the isomeric diphenyloxetanes,[1] the *cis* isomer, **12**, has a lower R_f value compared with the *trans* isomer, **13**, on a silica gel plate with hexane-ethyl acetate. Furthermore, the C-2 hydrogen in compound **13** appears at higher field (δ 5.78) in comparison with compound **12** (δ 6.20). Moreover, in compound **12**, the C-2 hydrogen (at δ 6.20) exhibits NOE effect caused by the proximity of the C-3 hydrogen. No such effect is observed with isomer **13**.

Methyl 4-[3-(3,5-dimethoxy-phenyl)-oxetan-2-yl]-benzoate, 6, 7. Compound **12** (60 mg) in methanol-THF (1:1, 4 mL) was treated with aq. NaOH (1M, 2.5 mL) for 16 h. The reaction was cooled with an ice-water bath, diluted with ethyl acetate, acidified with aq. HCl (1N), and extracted with ethyl acetate. The combined organic layer was washed with brine, dried over Na_2SO_4 and the solvent was removed under reduced pressure. The crude residue was filtered over silica gel (hexane-ethyl acetate) to afford compound **6** (45 mg). ^{1}H NMR ($CDCl_3$, 200 MHz): δ 7.92 (d, J = 8.0 Hz, 2H), 7.21 (d, J = 8.0 Hz, 2H), 6.23 (d, J = 8.5 Hz, 1H), 6.16 (s, 3H), 5.17 (t, J = 7.2 Hz, 1H), 4.86 (t, J = 6.3 Hz, 1H), 4.53 (q, J = 7.4 Hz, 1H), 3.59 (s, 6H).

In a similar manner, compound **13** (60 mg) was hydrolyzed to afford acid **7** (42 mg). ^{1}H NMR ($CDCl_3$, 200 MHz): δ 8.14 (d, J = 8.0 Hz, 2H), 7.52 (d, J = 8.0 Hz, 2H), 6.56 (d, J = 2.0 Hz, 2H), 6.40 (s, 1H), 5.85 (d, J = 6.9 Hz, 1H), 4.96 (m, 2H), 3.97 (m, 1H), 3.80 (s, 6H).

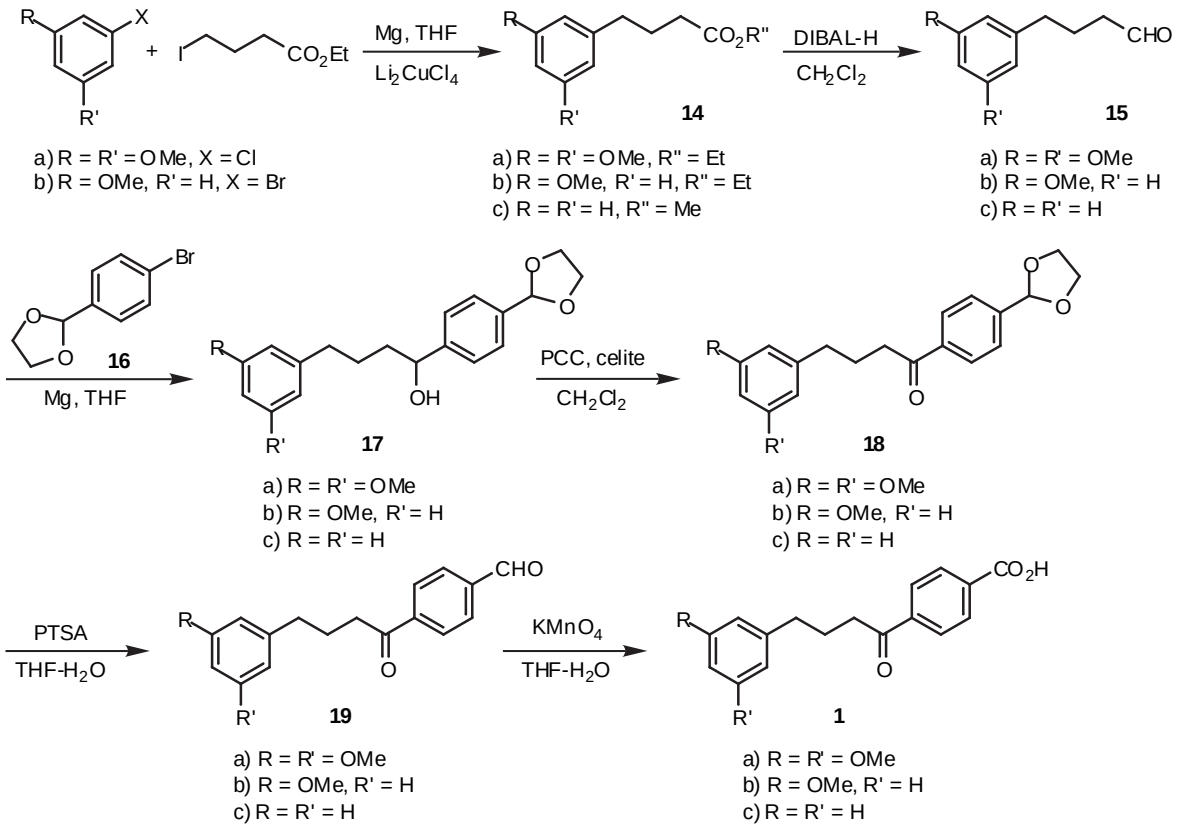

Figure 2: Synthesis of substrates 1a-c.

Ethyl 4-(3,5-dimethoxyphenyl)butyrate, 14a.[2] An oven dried three-necked flask was loaded with Mg (2.82 g, 0.116 mol) and a small volume of dry THF. The Mg was activated by the addition of a few drops of dibromoethane. Solution of 5-chloro-1,3-dimethoxybenzene (10 g, 0.058 mol) in THF (20 mL) was added dropwise and the mixture was refluxed for 6 h. A mixture of ethyl 4-iodobutyrate (14 g, 0.058 mol) and Li_2CuCl_4 (0.1M in THF, 14.4 mL) was prepared under argon and cooled to 0 ^{0}C. The above described Grignard solution was added via syringe over 30 min, the mixture was stirred at 0 ^{0}C for 30 min and then allowed to warm slowly to room temperature overnight. The mixture was acidified with HCl (6M, 20 mL) and extracted with diethyl ether (3x30 mL). The organic layer was washed with 15% aq. NH_4OH (40 mL), water (30 mL) and brine (30 mL). The combined organic layer was dried over Na_2SO_4, solvents were removed under reduced pressure and the residue was purified over two consecutive silica gel columns. (hexane:ethyl acetate 9:1) followed by another column (benzene) to give compound **14a**

in the form of a yellowish oil (4.16 g, 30%). ^{1}H-NMR (200 MHz, $CDCl_3$): 6.32 (d, J = 1.9 Hz, 2H), 6.29 (t, J = 1.9 Hz, 1H), 4.11 (q, J = 7.1 Hz, 2H), 3.76 (s, 6H), 2.57 (t, J = 7.6 Hz, 2H), 2.30 (t, J = 7.4 Hz, 2H), 1.92 (quintet J = 7 Hz, 2H), 1.23 (t, J = 7.1 Hz, 3H); ^{13}C-NMR (200 MHz, $CDCl_3$): 173.5, 160.8, 143.8, 106. 6, 98.0, 60.2, 55.2, 35.4, 33.6, 26.3, 14.2 ppm. MS (EI): 252 (M^+).

4-(3,5-Dimethoxy-phenyl)butyraldehyde, 15a. DIBAL-H (1.5 M in toluene, 4.4 mL, 6.6 mmol) was added dropwise to a solution of **14a** (1.34 g, 5.32 mmol) in dry CH_2Cl_2 (20 mL) at –78 ^{0}C. After stirring for 1 h, the mixture was quenched with saturated aq. NH_4Cl (1.4 mL), celite (1 g) was added, the mixture was diluted with diethyl ether (10 mL) and the temperature was raised to 0 ^{0}C. After stirring for an additional 0.5 h, more diethyl ether was added, the mixture was dried over Na_2SO_4 and filtered, solvents were removed under reduced pressure and the residue was purified over silica gel (hexane-ethyl acetate from 8:2 to 7:3) to give **15a** (777 mg, 70%) in the form of a colorless oil. ^{1}H-NMR (200 MHz, $CDCl_3$): 9.73 (t, J = 1.6 Hz, 1H), 6.30 (s, 3H), 3.76 (s, 6H), 2.58 (t, J = 7.5 Hz, 2H), 2.44 (t, J = 7.6 Hz, 2H), 1.85-2.00 (quintet, J = 7.5 Hz, 2H). MS (CI): 209.1 (MH^+).

4-(3,5-Dimethoxyphenyl)-1-[4-(dioxolan-2-yl)phenyl]butan-1-ol, 17a. An oven dried three-necked flask was loaded with Mg (300 mg, 12.5 mmol). The surface of the Mg was covered with dry THF and was activated by few drops of dibromoethane. Bromide **16** (available from TCI America) was added dropwise and the progress of the reaction was followed by GC. The solution was warmed to 40^{0}C and stirred for 40 min, then cooled to room temperature. Aldehyde **15a** (527 mg, 2.53 mmol) was dissolved in THF (10 mL) and cooled to 0 ^{0}C. The Grignard reagent solution was slowly added via a syringe. The mixture was stirred at 0 ^{0}C for 15 min and then allowed to warm to room temperature and stirred for additional 30 min. The reaction was quenched with saturated aq. NH_4Cl and extracted with diethyl ether. The combined organic phase was washed with brine, dried over $MgSO_4$. Solvents were removed under reduced pressure. The crude product was purified over silica gel (hexane-ethyl acetate 8:2) to give **17a** (580 mg, 64%). ^{1}H NMR (200 MHz, $CDCl_3$): 7.43 (d, J = 8.3 Hz, 2H), 7.31 (d, J = 8.3 Hz, 2H), 6.30 (m, 3H) 5.78 (s, 1H), 4.67 (t, J = 6.9 Hz, 1H), 3.97-4.15 (m, 4H), 3.74 (s, 6H), 2.53 (t, J = 7.2 Hz, 2H), 1.56-1.89 (m, 4H).

4-(3,5-Dimethoxyphenyl)-1-[4-(dioxolan-2-yl)phenyl]butan-1-one, 18a. PCC (1.99 g, 9.2 mmol) and celite (2 g) were added to a stirring solution of alcohol **17a** (1.65 g, 4.6 mmol) in CH_2Cl_2 (20 mL) at room temperature. After 1 h the brown mixture was filtered through silica gel and washed with ethyl acetate. The solvents were removed under reduced pressure and the crude product was purified by column chromatography (silica gel, hexane-ethyl acetate 7:3) to give **18a** (1.17 g, 72%). ^{1}H NMR (200 MHz, $CDCl_3$): 7.91 (d, J = 8.2 Hz, 2H), 7.53 (d, J = 8.2 Hz, 2H), 6.33 (d, J = 2.2 Hz, 2H), 6.29 (t, J = 2.2 Hz, 1H), 5.78 (s, 1H)4.02-4.11 (m, 4H), 3.75 (s, 6H), 2.95 (t, J = 7.2 Hz, 2H), 2.64 (t, J = 7.5 Hz, 2H), 2.05 (quintet, J = 7.3 Hz, 2H). MS (CI): 357.2 (MH^+).

4-(3,5-Dimethoxyphenyl)-1-(4-formylphenyl)-butan-1-one, 19a. Compound **18a** (248 mg, 0.7 mmol) was dissolved in THF (5 mL) and H_2O (0.5 mL). PTSA (50 mg) was added and the mixture was stirred over night. Saturated aq. $NaHCO_3$ was added and the mixture was extracted with diethyl ether. The combined organic phase was washed with brine, dried over $MgSO_4$ and solvents were removed under reduced pressure. The crude product was purified by column chromatography (silica gel, hexane-ethyl acetate 7:3) to give **19a** (180 mg, 78.4%) in the form of a white solid. ^{1}H NMR (200 MHz, $CDCl_3$): 10.07 (s, 1H), 8.03 (d, J = 8.2 Hz, 2H), 7.93 (d, J = 8.2 Hz, 2H), 6.33 (d, J = 1.7 Hz, 2H), 6.30 (t, J = 1.7 Hz, 1H), 3.75 (s, 6H), 2.99 (t, J = 7.1 Hz, 2H), 2.66 (t, J = 7.4 Hz, 2H), 2.07 (quintet, J = 7.3, 2H). MS (CI): 313.2 (MH^+).

4-(3,5-Dimethoxyphenyl)-1-(4-hydroxycarbonylphenyl)-butan-1-one, 1a. A solution of **19a** (97 mg, 0.31 mmol) in *t*-BuOH (2.3 mL) was diluted with aq. potassium phosphate buffer (1.25 M pH 4, 1.5 mL). Aq. $KMnO_4$ (1 M, 2.3 mL) was added with vigorous stirring, the mixture was stirred at room temperature for 10 minutes and then quenched by the addition of saturated aq. Na_2SO_3. The resultant pH of the mixture was adjusted to 3 with dilute HCl, and extracted with diethyl ether The combined organic phase was washed with brine, dried over $MgSO_4$ and solvents were removed under reduced pressure. The crude product was purified by column chromatography (silica gel, hexane-ethyl acetate 7:3 + 0.5% acetic acid) to give **1** (52 mg, 51%) in the form of a white solid. ^{1}H NMR (200 MHz, $CDCl_3$): 8.15 (d, J = 8.3 Hz, 2H), 7.96 (d, J = 8.3 Hz, 2H), 6.33 (br s, 2H), 6.30 (br s, 1H), 3.75 (s, 6H), 3.99 (t, J = 7.1 Hz, 2H), 2.66 (t, J = 7.4 Hz, 2H), 2.09 (quintet, J = 7.1 Hz, 2H). MS (CI): 328 (M^-).

Physical data of similarly prepared analogs:

Ethyl 4-(3-Methoxyphenyl)butyrate, 14b.[3] This analog was prepared from 3-methoxy-1-bromobenzene). ^{1}H NMR (200 MHz, $CDCl_3$): 7.22-7.14 (m, 1H), 6.77-6.71 (m, 3H), 4.10 (q, J = 7.4 Hz, 2H), 3.78 (s, 3H), 2.61 (t, J = 7.3 Hz, 2H), 2.28 (t, J = 7.3 Hz, 2H), 1.97 (quintet, J = 7.3 Hz, 2H), 1.23 (t, J = 7.4 Hz, 3H). MS (CI): 223.2 (MH^+).

4-(3-Methoxy-phenyl)butyraldehyde, 15b.[4] ^{1}H NMR (200 MHz, $CDCl_3$): 9.74 (br s, 1H), 7.19 (br t, J = 7.8 Hz, 1H), 6.70 (m, 3H), 3.78 (s, 3H), 2.62 (t, J = 7.4 Hz, 2H), 2.44 (t, J = 7.7 Hz, 2H), 1.94 (quintet, J = 7.5 Hz, 2H). MS (CI): 179.1 (MH^+).

4-(3-Methoxyphenyl)-1-[4-(dioxolan-2-yl)phenyl]butan-1-ol, 17b. ^{1}H NMR (200 MHz, $CDCl_3$): 7.43 (d, J = 8.2 Hz, 2H), 7.31 (d, J = 8.2 Hz, 2H), 6.73-6.68 (m, 4H), 5.78 (s, 1H), 4.67 (t, J = 6.9 Hz, 1H), 4.15-4.00 (m, 4H), 3.76 (s, 3H), 2.57 (t, J = 7.3 Hz, 2H), 1.88-1.75 (m, 4H). MS (CI): 329.0 (MH^+).

4-(3-Methoxyphenyl)-1-[4-(dioxolan-2-yl)phenyl]butan-1-one, 18b. ^{1}H NMR (200 MHz, $CDCl_3$): 7.91 (d, J = 8.2 Hz, 2H), 7.53 (d, J = 8.2 Hz, 2H), 7.18 (t, J = 8.2 Hz, 1H), 6.79-6.71 (m, 3H), 5.83 (s, 1H), 4.17-4.02 (m, 4H), 3.77 (s, 3H), 2.96 (t, J = 7.2 Hz, 2H), 2.68 (t, J = 7.3 Hz, 2H), 2.03 (quintet, J = 7.3 Hz, 2H). MS (CI): 327.0 (MH^+).

4-(3-Methoxyphenyl)-1-(4-formylphenyl)-butan-1-one, 19b. ^{1}H NMR (200 MHz, $CDCl_3$): 10.08 (s, 1H), 8.03 (d, J = 8.3 Hz, 2H), 7.93 (d, J = 8.3 Hz, 2H), 7.21-7.13 (m, 1H), 6.79-6.68 (m, 3H), 3.77 (s, 3H), 2.99 (t, J = 7.1 Hz, 2H), 2.70 (t, J = 7.1 Hz, 2H), 2.08 (quintet, J = 7.1 Hz, 2H). MS (CI): 283.1 (MH^+).

4-(3-Methoxyphenyl)-1-(4-hydroxycarbonylphenyl)-butan-1-one, 1b. ^{1}H NMR (200 MHz, $CDCl_3$): 8.15 (d, J = 8.4 Hz, 2H), 7.97 (d, J = 8.4 Hz, 2H), 7.17 (m, 1H), 6.79-6.69 (m, 3H), 3.77 (s, 3H), 3.00 (t, J = 7.2 Hz, 2H), 2.72 (t, J = 7.2 Hz, 2H), 2.08 (quintet, J = 7.2 Hz, 2H). MS (CI): 299.1 (MH^+).

Ethyl 4-phenylbutyrate, 14c. (prepared by the esterefication of 4-Phenyl-butyric acid) ^{1}H NMR (200 MHz, $CDCl_3$): 7.31-7.14 (m, 5H), 3.65 (s, 3H), 2.64 (t, J = 7.4 Hz, 2H), 2.32 (t, J = 7.3 Hz, 2H), 1.94 (quintet, J = 7.4 Hz, 2H). MS (CI): 179.0 (MH^+). MS (CI): 179.0 (MH^+).

4-Phenylbutyraldehyde, 15c.[5] ^{1}H NMR (200 MHz, $CDCl_3$): 9.74 (t, J = 1.5 Hz, 1H), 7.32-7.14 (m, 5H), 2.65 (t, J = 7.3 Hz, 2H), 2.44 (td, J = 7.7 Hz, 1.4 Hz, 2H), 1.95 (quintet, J = 7.7 Hz, 2H). MS (CI): 149 (MH^+).

4-Phenyl-1-[4-(dioxolan-2-yl)phenyl]butan-1-ol, 17c. ^{1}H NMR (200 MHz, $CDCl_3$): 7.45 (d, J = 8.2 Hz, 2H), 7.33 (d, J = 8.2 Hz, 2H), 7.26-7.12 (m, 5H), 5.80 (s, 1H), 4.70 (t, J = 6.9 Hz, 1H), 4.14-4.02 (m, 4H), 2.62 (t, J = 7.2 Hz, 2H), 1.79-1.63 (m, 4H). MS (CI): 299.2 (MH^+).

4-Phenyl-1-[4-(dioxolan-2-yl)phenyl]butan-1-one, 18c. ^{1}H NMR (200 MHz, $CDCl_3$): 7.90 (d, J = 8.3 Hz, 2H), 7.53 (d, J = 8.3 Hz, 2H), 7.32-7.16 (m, 5H), 5.83 (s, 1H), 4.15-3.99 (m, 4H), 2.95 (t, J = 7.1 Hz, 2H), 2.70 (t, J = 7.5 Hz, 2H), 2.06 (quintet, J = 7.1-7.5 Hz, 2H). MS (CI): 297.1 (MH^+).

4-Phenyl-1-(4-formylphenyl)-butan-1-one, 19c. ^{1}H NMR (200 MHz, $CDCl_3$): 10.08 (s, 1H), 8.03 (d, J = 8.2 Hz, 2H), 7.93 (d, J = 8.2 Hz, 2H), 7.33-7.15 (m, 5H), 2.99 (t, J = 7.2, 2H), 2.72 (t, J = 7.5 Hz, 2H), 2.08 (quintet, J = 7.4 Hz, 2H). MS (CI): 253.1 (MH^+).

4-Phenyl-1-(4-hydroxycarbonylphenyl)-butan-1-one, 1c. ^{1}H NMR (200 MHz, $CDCl_3$): 8.17 (d, J = 8.3 Hz, 2H), 7.96 (d, J = 8.3 Hz, 2H), 7.33-7.16 (m, 5H), 2.99 (t, J = 7.1 Hz, 2H), 2.72 (t, J = 7.1 Hz, 2H), 2.08 (quintet, J = 7.1 Hz, 2H). MS (CI): 269.1 (MH^+).

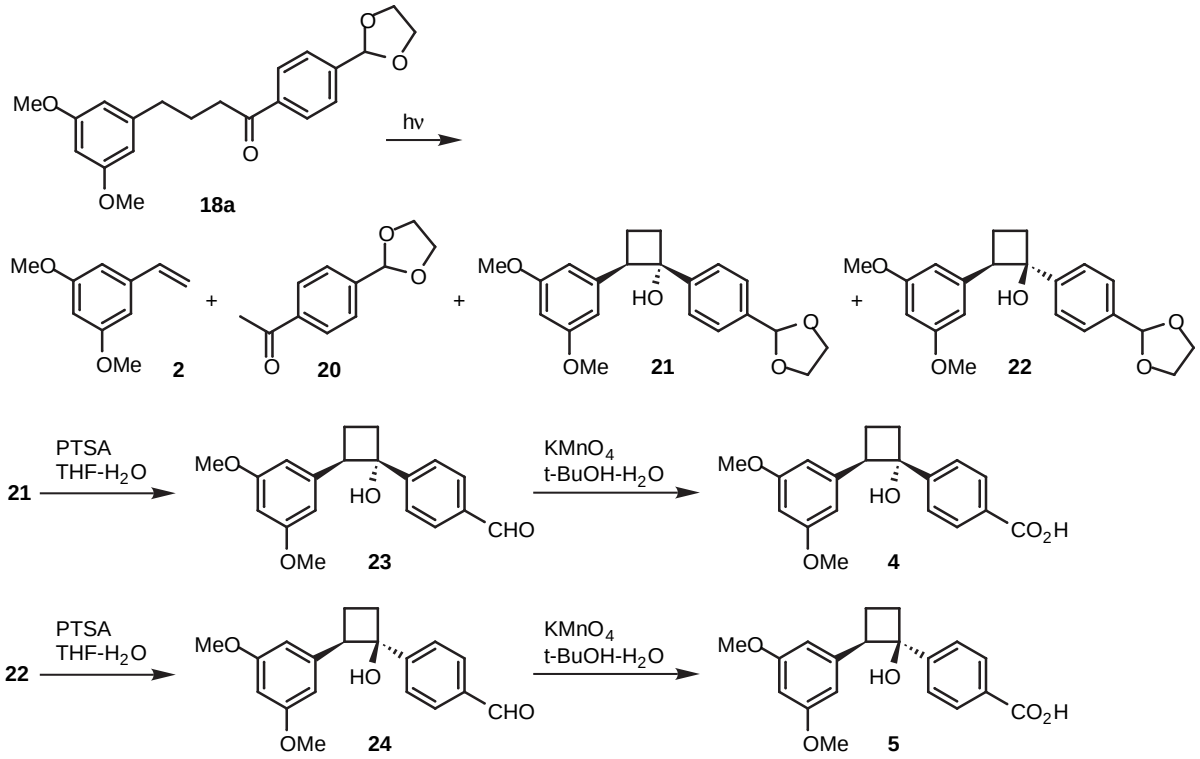

Figure 3: **Synthesis of cyclobutanols 4 and 5.** All structures represent relative, not absolute stereochemistry. Compounds **21, 23, 4** are refered to as cis (formal nomenclature gives E) and compounds **22, 24,5** are refered to as trans. See text for stereochemical assignments.

Irradiation of 18a. Ketone **18a** (676 mg, 1.98 mmol) was dissolved in acetone (10 mL) under argon, the solution was flushed with argon for 10 min and then irradiated with a mercury lamp for 7 h. The solvent was removed under reduced pressure and the residue was separated by flash column chromatography (silica gel, 2-10% ethyl acetate in benzene) to give **2** (127 mg, 41%), **20** (165 mg, 45.5%), **21** (33 mg, 4.9%), and **22** (35 mg, 5.2%)**.**

3,5-dimethoxystyrene, 2. ^{1}H NMR (200 MHz, $CDCl_3$): 6.61-6.70 (dd, J = 17.4, 10.8 Hz, 1H), 6.55 (d, J = 2.2 Hz, 2H), 6.37 (t, J = 2.2 Hz, 1H), 5.71 (d, J = 17.4 Hz, 1H), 5.23 (d, J = 10.8 Hz, 1H), 3.79 (s, 6H). MS (CI): 165.0 (MH^+).

4-(dioxolan-2-yl)acetophenone, 20. ^{1}H NMR (200 MHz, $CDCl_3$): 7.95 (d, J = 8.2 Hz, 2H), 7.55 (d, J = 8.2 Hz, 2H), 5.85 (s, 1H), 3.00-4.14 (m, 4H), 2.59 (s, 3H). MS (CI): 193.2 (MH^+).

***cis*-2-(3,5-Dimethoxyphenyl)-1-[4-(dioxolan-2-yl)phenyl]cyclobutan-1-ol, 21.** ^{1}H NMR (200 MHz, $CDCl_3$): 7.30 (d, J = 8.3 Hz, 2H), 7.24 (d, J = 8.3 Hz, 2H), 6.12 (t, J = 2.2 Hz, 1H), 5.96 (d, J = 2.2 Hz, 2H), 5.72 (s, 1H), 3.96-4.09 (m, 4H), 3.79 (dd, J = 10.3, 9.2, Hz, 1H), 3.57 (s, 6H), 2.70-2.78 (m, 1H), 1.86-2.41 (m, 3H). MS (EI): 356 (M^+).

***trans*-2-(3,5-Dimethoxyphenyl)-1-[4-(dioxolan-2-yl)phenyl]cyclobutan-1-ol, 22.** ^{1}H NMR (200 MHz, $CDCl_3$): 7.52 (d, J = 8.7 Hz, 2H), 7.46 (d, J = 8.7 Hz, 2H), 6.33 (d, J = 1.9 Hz, 2H), 6.30 (t, J = 1.9 Hz, 1H), 5.80 (s, 1H), 3.98-4.15 (m, 4H), 3.90 (t, J = 8.0 Hz, 1H), 3.72 (s, 6H), 2.43-2.54 (m, 2H), 2.17-2.30 (m, 2H). MS (EI): 356 (M^+).

Determination of stereochemistry of the *cis*- and *trans*-cyclobutanol derivatives.

A literature report on the structure of *cis*- and *trans*-1,2-diphenylcyclobutanols was unequivocally supported by X-ray crystallography of the *cis* isomer.[6] The reported ^{1}H NMR data of these isomers differ significantly from one another, particularly in the region of 1.8-3.0 ppm, which represents the cyclobutanol methylene. Since the NMR spectra of our isomers, **21** and **22** matched the reported spectra of 1,2-diphenylcyclobutanols, the relative stereochemistry of our compounds was also determined unequivocally. This assignment was further confirmed by NOE experiments. Significant NOE effect was observed with the *trans* isomer, **22**. We carried out a 1D NOE experiment and used multiple selective frequency version of NOE, where each line of a multiplet was irradiated for a short period of time within a narrow bandwidth. The NOE experiment confirmed the assignment of the *trans* isomer (data not shown) with its benzylic hydrogen at 3.9 ppm exhibiting proximity to both aromatic rings, a situation that was not seen with the *cis* isomer. Our assumption that the less polar cyclobutanol (as seen on a TLC plate) was the *trans* isomer was thus confirmed by NMR spectroscopy.

***cis*-2-(3,5-Dimethoxyphenyl)-1-(4-formylphenyl)-cyclobutanol, 23.** Compound **21** (13 mg, 0.037 mmol) was dissolved in THF (9 mL) and H_2O (1 mL). PTSA (10 mg) was added and the mixture was stirred overnight. A solution of saturated aq. $NaHCO_3$ was added and the mixture was extracted with diethyl ether. The combined organic phase was washed with brine, dried over $MgSO_4$, and solvents were removed under reduced pressure. The crude product was purified by flash column chromatography (silica gel, hexane-ethyl acetate 3:7) to give **23** (7 mg, 61.5%). ^{1}H

NMR (200 MHz, $CDCl_3$): 9.90 (s, 1H), 7.68 (d, J = 8.3 Hz, 2H), 7.43 (d, J = 8.3 Hz, 2H), 6.11 (t, J = 2.2 Hz, 1H), 5.98 (d, J = 2.2 Hz, 2H), 3.86 (t, J = 9.8 Hz, 1H), 3.58 (s, 6H), 2.79 (br t, J = 9.8 Hz, 1H), 1.97-2.43 (m, 3H). MS (CI): 313.2 (MH^+).

***trans*-2-(3,5-Dimethoxyphenyl)-1-(4-formylphenyl)-cyclobutanol, 24.** The synthesis of **24** from **22** was analogous to that of **23** from **21**. The crude product was purified by flash column chromatography (silica gel, hexane-ethyl acetate 2:8) to give **24** (24 mg, 77%). ^1H NMR (200 MHz, $CDCl_3$): 9.99 (s, 1H), 7.67 (d, J = 8.2 Hz, 2H), 7.66 (d, J = 8.2 Hz, 2H), 6.34 (t, J = 2.2 Hz, 1H), 6.25 (d, J = 2.1 Hz, 2H), 3.93 (t, J = 8.1 Hz, 1H), 3.72 (s, 6H), 2.55-2.66 (m, 2H), 2.26-2.35 (m, 2H). MS (CI): 313.2 (MH^+).

***cis*-2-(3,5-Dimethoxyphenyl)-1-(4-hydroxycarbonylphenyl)cyclobutanol, 4.** A solution of **23** (18 mg, 0.058 mmol) in *t*-BuOH (0.6 mL) was diluted with aq. potassium phosphate buffer (1.25 M, pH = 4, 0.3 mL) and treated with aq. $KMnO_4$ (1 M, 0.53 mL) at room temperature as described above for the conversion of **19a** to **1a**. Compound **4** was obtained (18.9 mg, 100%) in the form of a white solid. ^1H NMR (200 MHz, CD_3OD): 7.79 (d, J = 8.2 Hz, 2H), 7.38 (d, J = 8.2 Hz, 2H), 6.10 (t, J = 2.2 Hz, 1H), 5.99 (d, J = 2.2 Hz, 2H), 3.83 (t, J = 9.8 Hz, 1H), 3.56 (s, 6H), 2.76 (br t, J = 9.8 Hz, 1H), 1.97-2.43 (m, 3H).

***trans*-2-(3,5-Dimethoxyphenyl)-1-(4-hydroxycarbonylphenyl)cyclobutanol, 5.** The synthesis followed the same procedure as for compound **4**. The crude product was purified by column chromatography (silica gel, hexane-ethyl acetate 1:1) to give **5** (13.6 mg, 98%). ^1H NMR (200 MHz, $CDCl_3$): 8.10 (d, J = 8.4 Hz, 2H), 7.61 (d, J = 8.4 Hz, 2H), 6.35 (t, J = 2.1 Hz, 1H), 6.27 (d, J = 2.1, 2H), 3.93 (t, J = 8.4 Hz, 1H), 3.73 (s, 6H), 2.48-2.67 (m, 2H), 2.21-2.34 (m, 2H).

Wavelength dependence experiments. An Oriel Xe 300-1000W lamp model 6269 was used as the light source. An Acton Research Corporation Spectra Pro – 275 monochromator with 0.275 Meter triple grating was used. Light intensities were measured using an Ophir Nova laser star energy monitor with a p-type thermophile absorber head. A scale of relative intensities was constructed and sample was irradiated at each wavelength for the appropriate time, so that all samples would receive the same amount of irradiation and all results would remain at less then

20% conversion. The samples were placed at the monochromator light exit using Starna quartz cuvets with 1 mm cell length. Each sample consisted of 50 µL solution. Experiments with Antibodies were performed with equal concentration of antibody and substrate, 50µM in PBS (pH 7.4) and 10% CH_3CN. Background experiments were performed using the same conditions, omitting the antibody. All solutions were stored at 0 ˚C. Each sample was thawed and injected to HPLC. No change in results was observed whether the sample was injected immediately after irradiation or two days later. All experiments were carried out in duplicates.

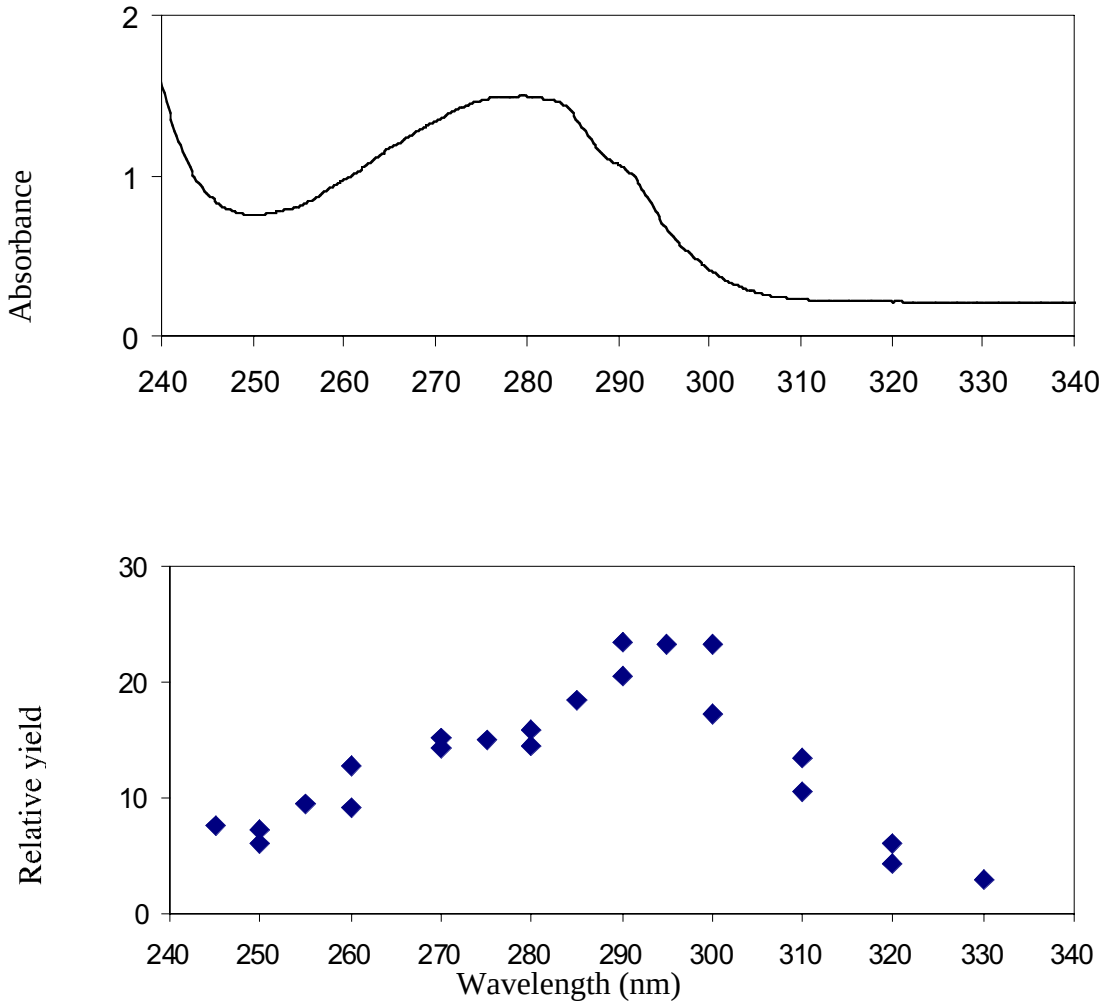

Figure 4. Top: absorption spectra of antibody 20F10. Bottom: relative yield of cyclization with antibody 20F10 as a function of wavelength.

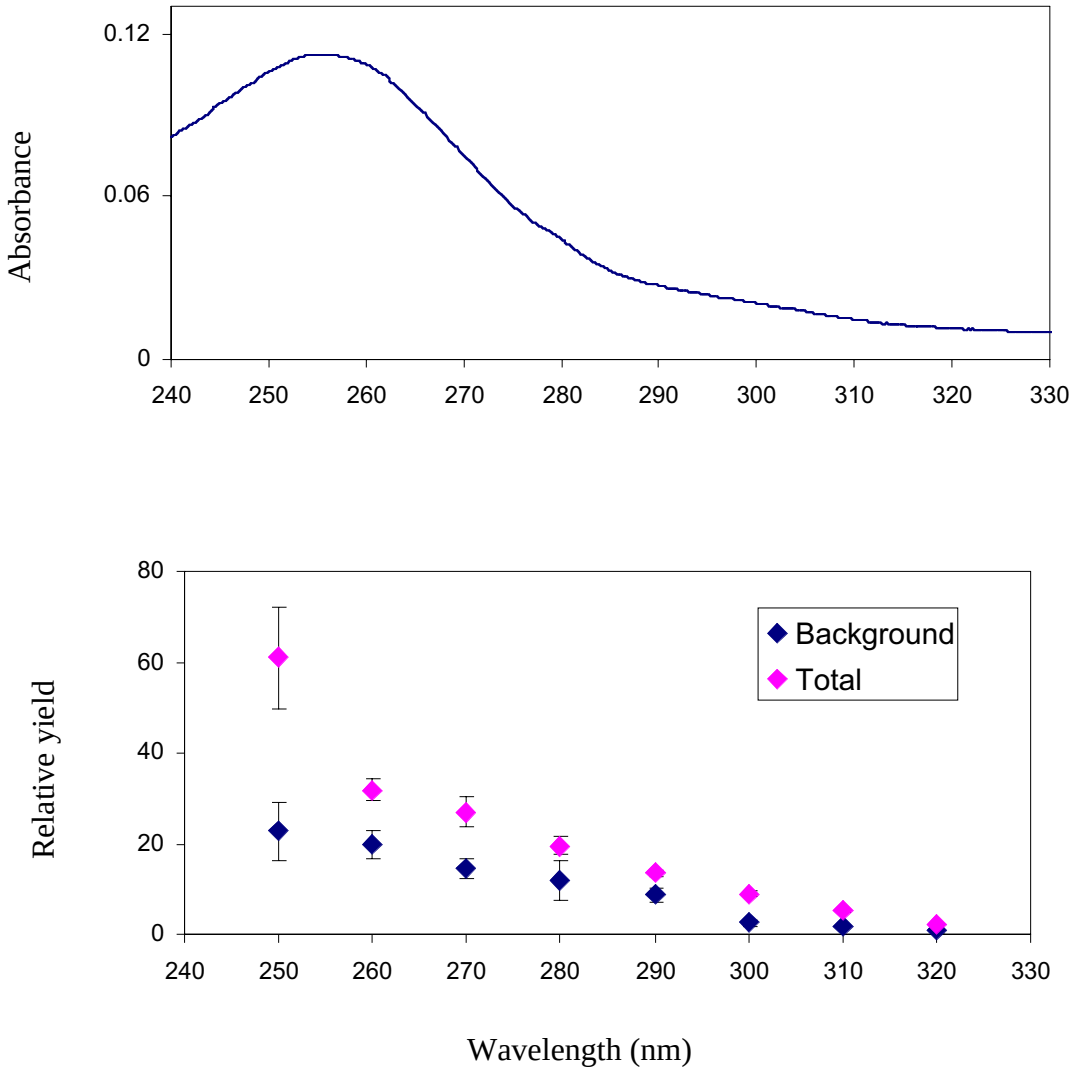

Figure 5. Top: absorption of substrate **1a** (10^{-5} M in PBS in the absence of antibody). Bottom: relative yield of fragmentation as a function of wavelength.

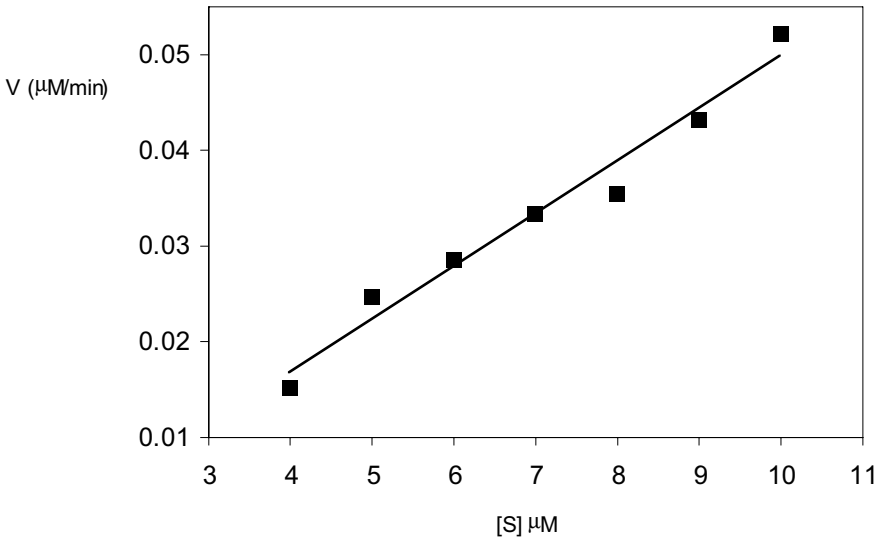

Figure 6. Initial rates of product formation under conditions of excess antibody give a pseudo-first-order rate constant: k_{obs}.

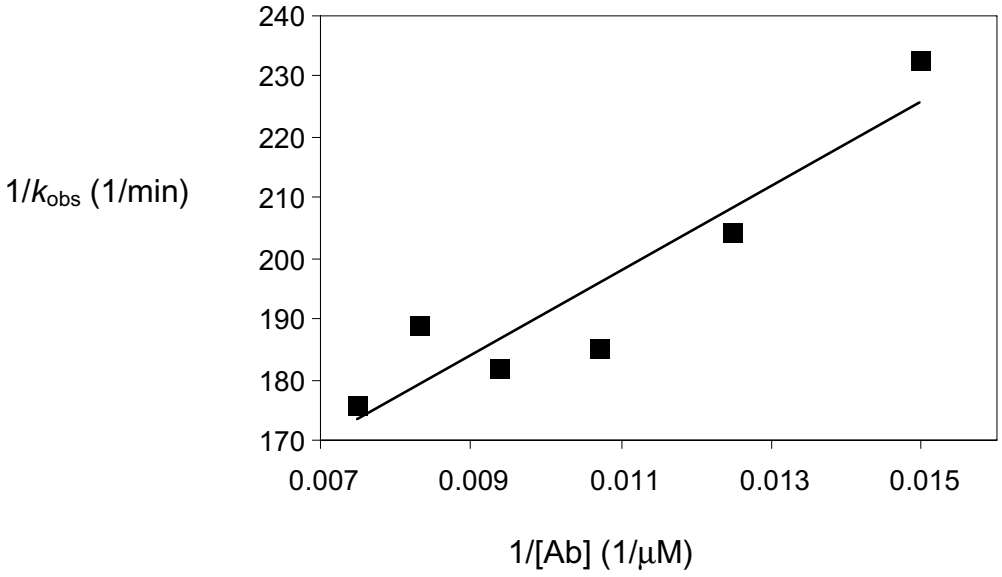

Figure 7. A double reciprocal plot of k_{obs} obtained by a series of kinetic measurements vs. antibody concentration, providing the values of k_{cat} and K_M.

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