Supplemental material for

Novel Cyclic Tripeptides and Substituted Aromatic Amino Acids Via Ruthenium Activated S_NAr Reactions.

Christopher W. West[1] and Daniel H. Rich[1,2]*

Department of Chemistry[1] and School of Pharmacy[2], University of Wisconsin-Madison, Madison, WI, 53706.

A. Materials and Methods

^{1}H NMR spectra were taken on a 300 MHz Bruker Aspect 3000 system. Chemical shifts were reported in ppm (δ units) downfield from tetramethylsilane (TMS) as the internal standard except for MeOH-d_4 and CD_3CN where the solvent peaks were used as internal standards. Electron Impact mass spectra (EIMS) was determined on a Kratos MS-80RFA spectrometer. LSIMS mass spectra was determined on a VG Auto Spec spectrometer.

THF was distilled from Na/benzophenone. DMF was supplied by Aldrich Chemical Company in Sure-Seal bottles. All other solvents were purchased from Aldrich and used without further purification. All amino acids were purchased from Bachem California except for (DL) 3-Chloro-phenylalanine and (DL) 4-Chloro-phenylalanine from Lancaster. All other reagents were purchased from Aldrich. All amino acids are L(S) designation unless noted otherwise.

All reactions were carried out under an atmosphere of argon using flame or oven dried glassware. A Buchi rotary evaporator equipped with a water condenser, a dry ice trap, and a water aspirator was used for the concentrated *in vacuo* steps.

Liquid chromatography was performed by flash chromatography using Merck grade 60 silica gel (230-400 mesh). Analytical thin layer chromatography (TLC) was run on Merck Kiesselgel $60F_{254}$ precoated plates of 0.25 mm thickness. For visualization, ultraviolet light and polyphosphomolybdic acid in EtOH or ninhydrin in EtOH with H_2SO_4 was used. The alumina used was activated, neutral Brockman I-grade (150 mesh).

B. Abbreviations

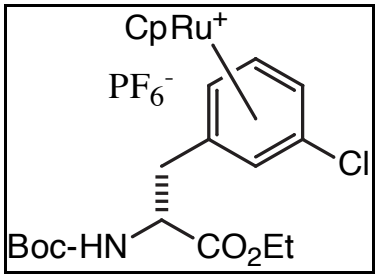
Boc	*tert*-butoxycarbonyl
Cp	cyclopentadiene
DCE	1, 2-dichloroethane
DMF	N, N-dimethylformamide
EtOAc	ethyl acetate
Fm	fluorenylmethyl
K O^{t}Bu	potassium *tert*-butoxide
Mtt	4-methyltrityl
Mmt	4-methoxytrityl
MTBD	1, 3, 4, 6, 7, 8-hexahydro-1-methyl-2H-pyrimido[1, 2-a]pyrimidine
TASF	tris(dimethylamino)sulfur (trimethylsilyl)difluoride
TBAF	tetrabutylamonium fluoride
Teoc	2-(trimethylsilyl)ethoxycarbonyl
THF	tetrahydrofuran

C. General Procedures

General Procedure A: Ruthenium Decomplexation

The ruthenium complexed starting material was dissolved in CH_3CN and degassed with Ar for 30 min. The reaction tube was then fitted with an Ar balloon and sparged. This was put in a Rayonet Photochemical Reactor and irradiated with 350 nm light for 24 hours. Upon completion, the reaction was concentrated *in vacuo*, redissolved in CH_2Cl_2, added to Et_2O to form a brown precipitate, and filtered. The product containing filtrate was concentrated *in vacuo* and purified by SiO_2 gel flash column chromatography.

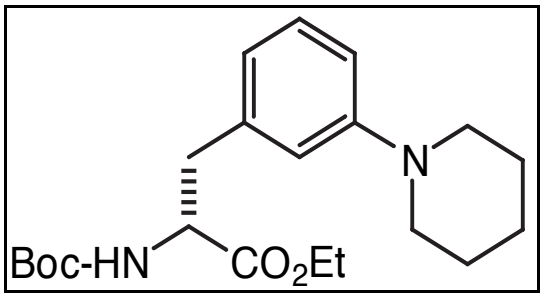
D. Specific Procedures



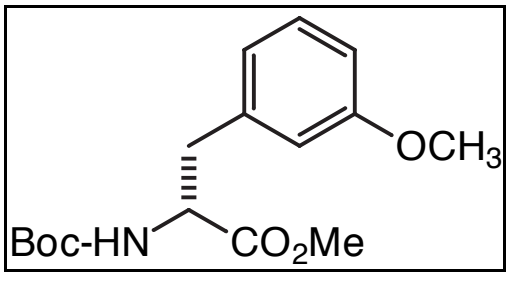
[η^6-(R)-(N-*tert*-Butoxycarbonyl)-3-chloro-phenylalanine ethyl ester](η^5-cyclopentadienyl)ruthenium hexafluorophosphate (3)

This is a typical procedure for complexation of ruthenium to an aromatic ring: A degassed solution of Boc (D) (3Cl) phenylalanine ethyl ester (144.8 mg, 0.44 mmol) in 4.0 mL of DCE was heated to reflux in an oil bath. Following addition of $RuCp(CH_3CN)_3PF_6$ (193.2 mg, 0.45 mmol) the solution immediately turned green and then changed to orange within 10 min. The reaction was refluxed for 2 hrs, allowed to cool, and concentrated *in vacuo* to give a brown foam. This was dissolved in CH_2Cl_2 and filtered through a short pad of neutral Al_2O_3 using 5% MeOH/CH_2Cl_2. The filtrate was concentrated *in vacuo*, redissolved in CH_2Cl_2, and added to Et_2O to form a precipitate which was filtered and washed with Et_2O (2x) to give 267.6 mg (95%) of the product as a light brown solid. ^1H NMR (300 MHz, CD_3CN) δ 6.59-6.46(m, 2H), 6.18-5.98(m, 2H), 5.76(m, 1H), 5.40(s,

5H), 4.36(m, 1H), 4.16(q, 2H, J=7.0 Hz), 3.06(dd, 1H, J=14.0, 5.2 Hz), 2.81(m, 1H), 1.35(s, 9H), 1.23(t, 3H, J=7.0 Hz). MS calculated for (M^+) $C_{21}H_{27}NO_4ClRu$=493.98; found (LSIMSMS), 494.2(M^+).

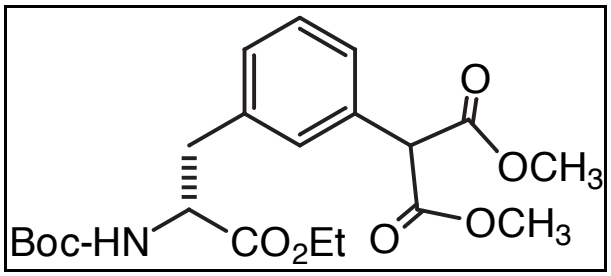


(R)-(N-*tert*-Butoxycarbonyl)-3-(piperidino) phenylalanine ethyl ester (4b) Dissolved **4a** (118.7 mg, 0.17 mmol) in 15 mL of CH_3CN and irradiated for 24 hours according to General Procedure A. The resulting brown solid was purified by flash column chromatography (SiO_2, 25% EtOAc/hexanes, 10 x 160 mm) to give 46.9 mg (72%) of product as a clear oil (R_f = 0.55, 40% EtOAc/hexanes). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.15(t, 1H), 6.81(dd, 1H, J=8.5, 2.2 Hz), 6.67(s, 1H), 6.58(d, 1H, J=7.4 Hz), 4.95(d, 1H, J=8.1 Hz), 4.53(m, 1H), 4.15(q, 2H, J=7.0 Hz), 3.13(t, 4H), 3.04(d, 2H, J=5.9 Hz), 1.69(m, 4H), 1.55(m, 2H), 1.42(s, 9H), 1.24(t, 3H, J=7.0 Hz); ^{13}C NMR (300 MHz, $CDCl_3$) δ 172.02, 155.11, 152.34, 136.68, 129.10, 120.03, 117.46, 115.11, 79.75, 61.25, 54.40, 50.57, 38.48, 28.32, 25.83, 24.31, 14.15. MS calculated for (M+Na) $C_{21}H_{32}N_2O_4$=399.2260; found (LSIMSMS), 399.2278 (M+Na).



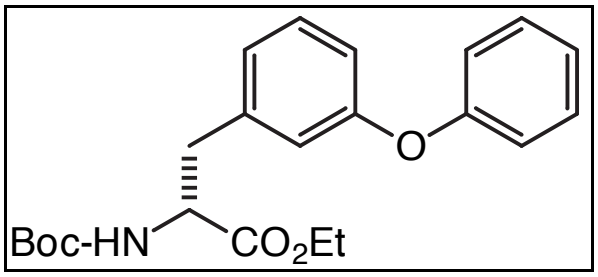
(R)-(N-*tert*-Butoxycarbonyl)-3-(methoxy) phenylalanine methyl ester (5b)

Dissolved **5a** (66.2 mg, 0.11 mmol) in 20 mL of CH_3CN and irradiated for 24.5 hours according to General Procedure A. The resulting brown solid was purified by flash column chromatography (SiO_2, 20% EtOAc/hexanes, 10 x 180 mm) to give 24.3 mg (70%) of product as a clear oil (R_f = 0.46, 40% EtOAc/hexanes). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.20(t, 1H), 6.79(dd, 1H, J=7.4, 2.6 Hz), 6.71(d, 1H, J=7.4 Hz), 6.67(s, 1H), 4.98(d, 1H, J=7.7 Hz), 4.57(m, 1H), 3.79(s, 3H), 3.72(s, 3H), 3.06(m, 2H), 1.42(s, 9H); ^{13}C NMR (300 MHz, $CDCl_3$) δ 172.30, 159.67, 137.50, 129.58, 129.52, 121.58, 114.95, 112.46, 79.91, 55.12, 54.34, 52.19, 38.29, 28.28. MS calculated for (M+Na) $C_{16}H_{23}NO_3$=332.1474; found (LSIMSMS), 332.1471(M+Na).

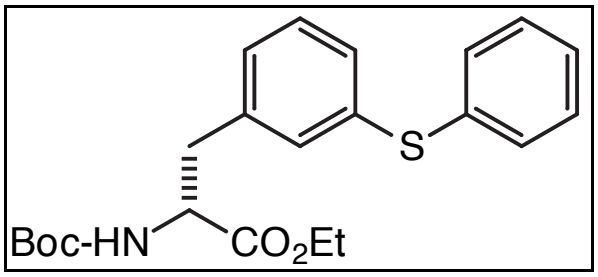


(R)-(N-*tert*-Butoxycarbonyl)-3-(dicarbomethoxymethyl) phenylalanine ethyl ester (6b)

Dissolved **6a** (82.5 mg, 0.11 mmol) in 50 mL of CH_3CN and irradiated for 24 hours according to General Procedure A. The resulting brown oil was purified by flash column chromatography (SiO_2, 30% EtOAc/hexanes, 10 x 200 mm) to give 26.3 mg (55%) of product as a clear oil (R_f = 0.39, 40% EtOAc/hexanes). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.28(m, 2H), 7.16(s, 1H), 7.1(m, 1H), 4.98(d, 1H, J=7.0 Hz), 4.61(s, 1H), 4.56(m, 1H), 4.15(q, 2H, J=7.0 Hz), 3.75(s, 6H), 3.10(d, 2H, J=5.5 Hz), 1.43(s, 9H), 1.23(t, 3H, J=7.0 Hz); ^{13}C NMR (300 MHz, $CDCl_3$) δ 171.68, 168.42, 155.09, 136.62, 132.75, 130.26, 129.41, 128.82, 127.93, 79.90, 61.41, 57.44, 54.32, 52.87, 38.13, 28.29, 14.08. MS calculated for (M+Na) $C_{21}H_{29}NO_8$=446.1791; found (LSIMSMS), 466.1803(M+Na).

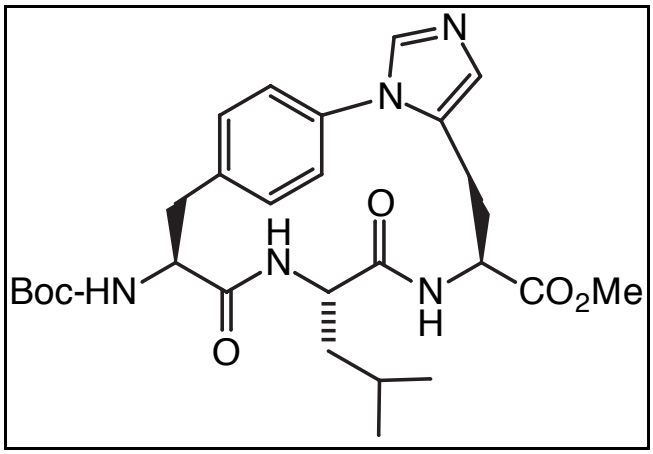


(R)-(N-*tert*-Butoxycarbonyl)-3-(phenoxy) phenylalanine ethyl ester (7b) Dissolved **7a** (50.3 mg, 0.072 mmol) in 10 mL of CH_3CN and irradiated for 22.5 hours according to General Procedure A. The resulting brown solid was purified by flash column chromatography (SiO_2, 40% EtOAc/hexanes, 10 x 120 mm) to give 17.6 mg (63%) of product as a clear oil (R_f = 0.70, 40% EtOAc/hexanes). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.35(t, 2H, J=8.1 Hz), 7.25(t, 1H, J=7.7 Hz), 7.10(t, 1H, J=7.4 Hz), 6.99(d, 2H, J=7.7 Hz), 6.88(m, 2H), 6.77(s, 1H), 5.00(d, 1H, J=7.7 Hz), 4.53(m, 1H), 4.10(m, 2H), 3.05(m, 2H), 1.41(s, 9H), 1.21(t, 3H, J=7.0 Hz); ^{13}C NMR (300 MHz, $CDCl_3$) δ 171.62, 157.35, 157.06, 155.01, 138.02, 129.74, 124.26, 123.29, 119.67, 118.94, 117.43, 79.86, 61.36, 54.29, 53.40, 38.16, 28.27, 14.09. MS calculated for (M+Na) $C_{22}H_{27}NO_3$=408.1787; found (LSIMSMS), 408.1786(M+Na).



(R)-(N-*tert*-Butoxycarbonyl)-3-(phenylthio) phenylalanine ethyl ester (8b) Dissolved **8a** (51.4 mg, 0.072 mmol) in 20 mL of CH_3CN and irradiated for 24.5 hours according to General Procedure A. The resulting brown solid was purified by flash column chromatography (SiO_2, 20% EtOAc/hexanes, 10 x 170 mm) to give 18.0 mg

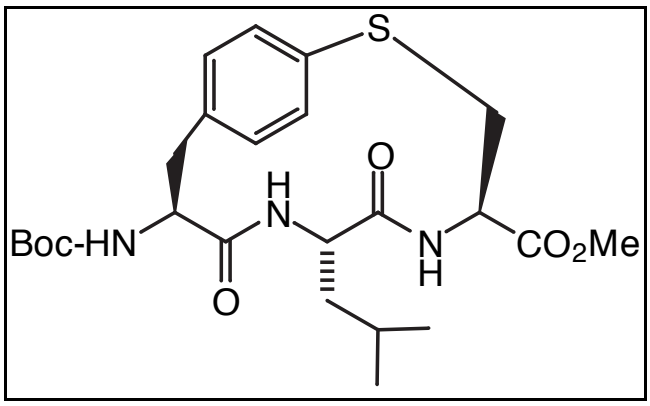
(62%) of product as a clear oil (R_f = 0.75, 5% MeOH/CH_2Cl_2). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.31(m, 5H), 7.26(m, 1H), 7.22(s, 1H), 7.13(s, 1H), 7.03(m, 1H), 4.98(d, 1H, J=8.1 Hz), 4.54(m, 1H), 4.10(m, 2H), 3.05(m, 2H), 1.42(s, 9H), 1.20(t, 3H, J=7.0 Hz); ^{13}C NMR (300 MHz, $CDCl_3$) δ 171.56, 155.00, 137.32, 135.86, 135.68, 131.99, 131.12, 129.66, 129.24, 129.19, 128.30, 127.04, 79.91, 61.39, 54.29, 38.11, 28.28, 14.11. MS calculated for (M+Na) $C_{22}H_{27}NO_3S$=424.1559; found (LSIMSMS), 424.1541(M+Na).



Cyclic (N-*tert*-Butoxycarbonyl)-*p*-phenylalanine leucine histidine methyl ester (17)

To a solution of **16** (72.4 mg, 0.083mmol) in 15 mL of THF and 2 mL DMF at -78˚C was added K O^{t}Bu (31.9 mg, 0.284 mmol). This was allowed to warm to room temp overnight and after a total reaction time of 72 hrs the solution was concentrated *in vacuo* to a brown oil. Ruthenium decomplexation took place over 26 hrs time using 40 mL of CH_3CN according to General Procedure A. The resulting oil was purified using flash column chromatography (SiO_2, 5% acetone/CH_2Cl_2, 10 x 240 mm) to give 10.3 mg (24%) of product (R_f = 0.22, 5% MeOH/CH_2Cl_2). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.97(s, 1H), 7.36(d, 1H, J=8.5 Hz), 7.18(d, 1H, J=8.8 Hz), 7.13(s, 2H), 6.59(s, 1H), 4.60(m, 1H), 3.97(m, 1H), 3.80(s, 4H), 3.32(m, 2H), 2.60(m, 2H), 1.53(m, 2H), 1.43(s, 9H),

1.35(m, 1H), 0.79(dd, 6H); ^{13}C NMR (300 MHz, $CDCl_3$) δ 172.01, 170.16, 169.64, 155.00, 138.46, 138.14, 136.69, 134.87, 131.75, 129.55, 125.05, 123.39, 120.56, 80.07, 58.30, 52.75, 51.90, 42.95, 38.96, 32.86, 28.26, 24.11, 22.97, 22.79. MS calculated for (M+Na) $C_{27}H_{37}N_5O_6$=550.2642, found (LSIMSMS) 550.2625(M+Na).



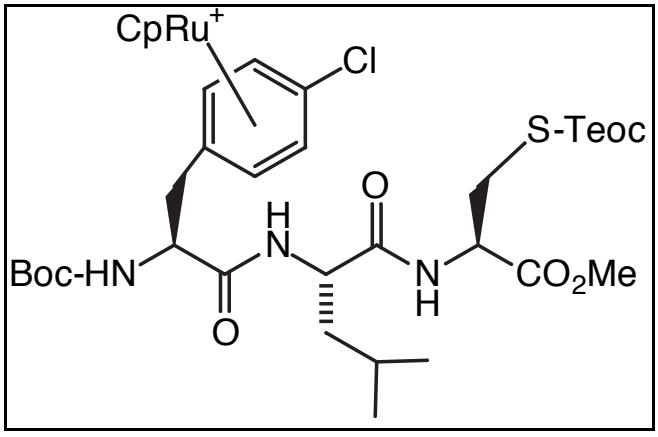
Cyclic (N-*tert*-Butoxycarbonyl)-*p*-phenylalanine leucine cysteine methyl ester (20)

From Mmt:

To a solution of Boc (4Cl) [(cyclopentadienyl)ruthenium hexafluorophosphate] phenylalanine leucine cysteine methyl ester (91.7 mg, 0.11 mmol) in 50 mL of THF and 5 mL of DMF at -78˚C was added 40% KF on Al_2O_3 (233.5 mg, 1.61 mmol) and 18-crown-6 (17.0 mg, 0.064 mmol). This was allowed to warm to room temp overnight and after a total reaction time of 6 days the solution was filtered, dried over $MgSO_4$, filtered, and concentrated *in vacuo* to yield a brown oil. Ruthenium decomplexation took place over 23 hrs time using 30 mL of CH_3CN according to General Procedure A. The resulting oil was purified using flash column chromatography (SiO_2, 20% EtOAc/hexanes, 10 x 180 mm) followed by rechromatography (SiO_2, 3% acetone/CH_2Cl_2, 10 x 240 mm) to give 9.8 mg (18%) of product as a white solid.

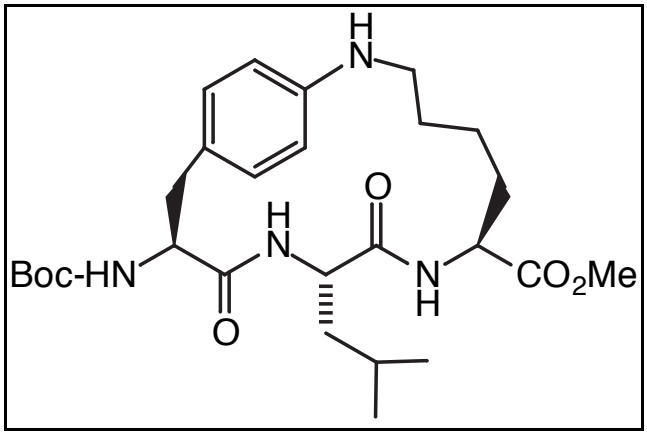
From Teoc:

To a solution of **23** (39.6 mg, 0.040 mmol) in 0.5 mL of THF and 0.5 mL of DMF at -78˚C was added 1.0 M in hexanes TBAF (0.08 mL, 0.08 mmol) and stirred for 18 hrs. The reaction was concentrated *in vacuo* to yield a brown oil. Ruthenium decomplexation took place over 19 hrs time using 30 mL of CH_3CN according to General Procedure A. The resulting oil was purified using flash column chromatography (SiO_2, 20% EtOAc/hexanes, 10 x 180 mm) followed by rechromatography (SiO_2, 3% acetone/CH_2Cl_2, 10 x 240 mm) to give 9.8 mg (52%) of product as a white solid (R_f = 0.32, 5% MeOH/CH_2Cl_2). ^{1}H NMR (300 MHz, DMSO-d_4) δ 7.40(d, 1H, J=8.1 Hz), 7.34(d, 1H, J=8.1 Hz), 7.24(m, 2H), 7.13(d, 1H, J=9.2 Hz), 7.02(d, 1H, J=7.4 Hz), 6.83(d, 1H, J=7.4 Hz), 4.61(m, 1H), 4.13(m, 1H), 3.83(m, 1H), 3.64(s, 3H), 3.34(hidden, 1H), 3.17(d, 1H, J=14.7 Hz), 3.00(dd, 1H, J=11.8, 6.6 Hz), 2.50(hidden, 1H), 1.37(s, 9H), 1.24(m, 2H), 1.13(m, 1H), 0.78(m, 6H); ^{13}C (300MHz, DMSO-d_4) δ 170.23, 170.02, 154.69, 137.22, 133.47, 133.37, 130.67, 130.18, 129.87, 77.91, 56.63, 55.96, 52.22, 50.43, 42.45, 37.73, 35.52, 28.11, 23.59, 22.61, 22.40. MS calculated for (M+Na) $C_{24}H_{35}N_3O_6S$=516.2144, found (LSIMSMS) 516.2167 (M+Na).



[η^6-(N-*tert*-butoxycarbonyl)-4-chloro-phenylalanine leucine cysteine (S-Teoc) methyl ester](η^5-cyclopentadienyl)ruthenium hexafluorophosphate (23)

To a degassed solution of **22** (96.0 mg, 0.14 mmol) in 8 mL of DCE at 75˚C was added $RuCp(CH_3CN)_3PF_6$ (64.1 mg, 0.15 mmol). The orange reaction was heated at reflux for 2 hrs and then allowed to cool. Removal of solvent under reduced pressure was followed by redissolving in CH_2Cl_2, adding to Et_2O to form a precipitate, and filtering to get 130.8 mg (93%) of product as a light tan solid. ^1H NMR (300 MHz, CD_3CN) δ 7.10(d, 1H, J=8.1 Hz), 6.96(d, 1H, J=7.4 Hz), 6.46(d, 2H, J=6.3 Hz), 6.16(t, 2H, J=6.3 Hz), 5.76(d, 1H, J=8.5 Hz), 5.38(s, 5H), 4.57(m, 1H), 4.28(m, 3H), 3.65(s, 3H), 3.36(dd, 1H, J=14.3, 5.2 Hz), 3.11(dd, 1H, J=14.3, 7.7 Hz), 2.94(dd, 1H, J=13.6, 5.2 Hz), 2.64(dd, 1H, J=13.6, 8.5 Hz), 1.89(m, 1H), 1.51(m, 2H), 1.33(s, 9H), 1.00(m, 2H), 0.87(m, 6H), 0.00(s, 9H). MS calculated for (M^+) $C_{35}H_{53}N_3O_8ClSSiRu$=840.5, found (LSIMSMS) 840.0(M^+).



Cyclic (N-*tert*-Butoxycarbonyl)-*p*-phenylalanine leucine lysine methyl ester (25)

From Mtt:

To a solution of Boc (4Cl) [(cyclopentadienyl)ruthenium hexafluorophosphate] phenylalanine leucine lysine (N-Mtt) methyl ester (143.8 mg, 0. mmol) in 75 mL of THF and 7 mL of DMF at -78˚C was added 40% KF on Al_2O_3 (413.8 mg, 2.85 mmol) and 18-crown-6 (29.2 mg, 0.11 mmol). This was allowed to warm to room temp overnight and

after a total reaction time of 77 hrs the solution was filtered, dried over $MgSO_4$, filtered, and concentrated *in vacuo* to a brown oil. Ruthenium decomplexation took place over 24 hrs using 30 mL of CH_3CN according to General Procedure A. The resulting oil was purified using flash column chromatography (SiO_2, 20% EtOAc/hexanes, 10 x 200 mm) followed by rechromatography (SiO_2, 2% MeOH/CH_2Cl_2, 10 x 220 mm) to give 2.0 mg (2%) of product as a white solid.

From Teoc:

To a solution of Boc (4Cl) [(cyclopentadienyl)ruthenium hexafluorophosphate] phenylalanine leucine lysine (N-Teoc) methyl ester (186.5 mg, 0.19 mmol) in 2.0 mL of THF and 1.8 mL of DMF was added TASF (150 mg, 0.54 mmol). The reaction was stirred at room temp for 62 hrs at which time the solvent was removed under reduced pressure to yield a brown oil that was dried *in vacuo* for 30 min. The brown oil was dissolved in 45 mL of CH_3CN and irradiated for 20 hrs according to General Procedure A. Removal of solvent was followed by redissolving in CH_2Cl_2, adding to Et_2O to form a precipitate, and filtering. The filtrate was concentrated *in vacuo*, filtered through a pad of SiO_2 using EtOAc and reconcentrated *in vacuo* to give an oil that was purified by flash column chromatography (SiO_2, 40% EtOAc/hexanes, 10 x 190 mm) to give 19.1 mg (20%) of product as a white solid (R_f = 0.37, 5% MeOH/CH_2Cl_2). ^{1}H NMR (300 MHz, $CDCl_3$/MeOH-d_4) δ 7.12(d, 2H, J=8.1 Hz), 6.87(d, 2H, J=8.1 Hz), 4.45(dd, 1H, J=11.0, 3.3 Hz), 4.25(dd, 1H, J=11.4, 5.5 Hz), 4.11(m, 1H), 3.70(s, 3H), 3.39(m, 1H), 3.24(m, 1H), 3.05(dd, 1H, J=12.9, 5.2 Hz), 2.94(t, 1H, J=12.5 Hz), 1.84(m, 1H), 1.69-1.40(m, 8H), 1.46(s, 9H), 1.21(m, 2H), 0.88(d, 6H, J=6.3 Hz); ^{13}C (300MHz, $CDCl_3$/MeOH-d_4) δ 172.34, 171.42, 170.25, 155.35, 137.58, 133.13, 130.45, 119.26, 80.00, 55.68, 52.03, 51.00, 50.79, 47.02, 42.12, 37.58, 30.43, 27.83, 25.30, 24.00, 22.97, 22.21, 21.95. MS calculated for (M+Na) $C_{27}H_{42}N_4O_6$=541.3002, found (LSIMSMS) 541.3007 (M+Na).