Supporting Information

Efficient Introduction of Protected Guanidines in BOC Solid Phase Peptide Synthesis

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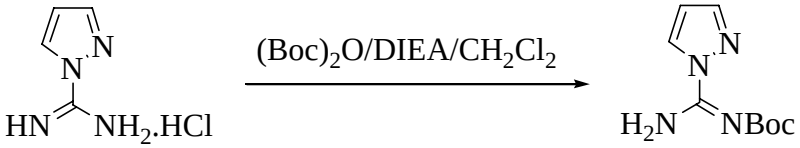
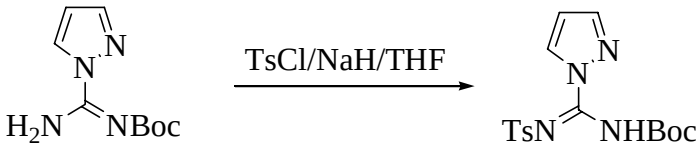
Synthesis of Reagent (1):



To a stirred solution of 1*H*-pyrazole-1-carboxamidine hydrochloride (5 g, 34.2 mmol) and DIEA (12 mL, 68.4 mmol) in 30 mL of dry CH_2Cl_2 was added a solution of $(Boc)_2O$ (7.1 g, 34.2 mmol) in 10 mL of CH_2Cl_2. After stirring at room temperature for 2 hours, 30 mL of CH_2Cl_2 was added and the mixture was washed with $NaHCO_3$ (sat.), water, and brine and dried over Na_2SO_4. After solvent removal under reduced pressure, the residue was chromatographed (3:1 hexane:ethyl acetate) to afford 6.5 g of the pure product (90 %).

^{1}H NMR (300 MHz, $CDCl_3$): δ 9.08 (s, 1H), 8.46 (d, *J* = 2.7 Hz, 1H), 7.67 (d, *J* = 1.2 Hz, 1H), 6.39 (dd, *J* = 1.2 Hz, *J* = 2.7 Hz, 1H), 1.54 (s, 9H).

^{13}C NMR (75 MHz, $CDCl_3$): δ 163.40, 155.13, 143.41, 128.90, 109.09, 80.36, 28.41.



To a suspension of NaH (1.7 g, 42.9 mmol) in 10 mL of THF was added dropwise a solution of the mono Boc derivative (3 g, 14.3 mmol) in 5 mL of THF at 0 ^{o}C. After the resulting mixture was stirred for half an hour at room temperature, a solution of TsCl (8.7g, 42.9 mmol) in 15 mL of THF was added. After further stirring for two hours at room temperature, water and CH_2Cl_2 were added and the organic layer was washed with $NaHCO_3$, water, brine and dried over Na_2SO_4. After solvent removal under reduced pressure, the residue was chromatographed (1:2 hexane:ethyl acetate) to yield 2.4g of the pure product (yield 87 % based on 1.4 g of recovered starting material). m.p. 139-141ºC

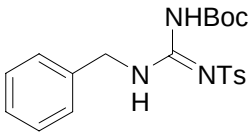
^{1}H NMR (300 MHz, $CDCl_3$): δ 8.95 (s, 1H), 8.08 (d, *J* = 2.7 Hz, 1H), 7.88 (d, *J* = 8.4 Hz, 2H), 7.74 (d, *J* = 1.2 Hz, 1H), 7.32 (d, *J* = 8.4 Hz, 2H), 6.43 (dd, *J* = 1.2 Hz, *J* = 2.7 Hz, 1H), 2.44 (s, 3H), 1.51 (s, 9H).

^{13}C NMR (75 MHz, $CDCl_3$): δ 148.78, 145.57, 144.60, 143.97, 138.40, 130.78, 129.70, 127.00, 110.47, 84.82, 30.01*, 28.03, 21.90.

HRMS: *m/z* calculated for $C_{16}H_{21}N_4O_4S$ [M+H] = 365.1284; found = 365.1289.

IR (cm^{-1}): 3119.84, 2981.66, 2930.77, 1761.39, 1620.36, 1154.33, 1089.50, 763.04, 668.23

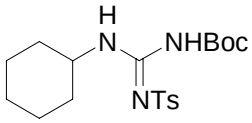
Characterization Data for Solution Products:



^{1}H NMR (300 MHz, $CDCl_3$, ppm): δ 9.97 (bs, 1H), 8.81 (bs, 1H), 7.73 (d, *J* = 8.1 Hz, 2H), 7.29 – 7.20 (m, 7H), 4.51 (d, *J* = 5.4 Hz, 2H), 2.41 (s, 3H), 1.49 (s, 9H)

^{13}C NMR (300 MHz, $CDCl_3$, ppm): δ 152.61, 150.88, 142.50, 140.41, 136.86, 129.35, 128.83, 127.93, 127.82, 126.07, 84.54, 45.41, 30.0*, 28.26, 21.79.

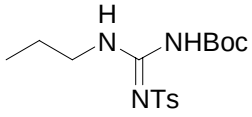
HRMS: *m/z* calculated for $C_{20}H_{26}N_3O_4S$ [M+H] = 404.1644; found = 404.1647.



^{1}H NMR (300 MHz, $CDCl_3$): δ 9.92 (s, 1H), 8.43 (d, *J* = 7.5 Hz, 1H), 7.78 (d. *J* = 8.4 Hz, 2H), 7.27 (d, *J* = 8.4 Hz, 2H), 3.88 – 3.79 (m, 1H), 2.41 (s, 3H), 1.91 – 1.14 (m, 19H).

^{13}C NMR (75 MHz, $CDCl_3$): δ 152.74, 149.97, 142.347, 140.75, 129.36, 126.00, 84.26, 50.00, 32.65, 30.0*, 28.31, 25.69, 24.67, 21.79.

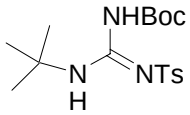
HRMS: *m/z* calculated for $C_{19}H_{30}N_3O_4S$ [M+H] = 396.1956; found 396.1957



^{1}H NMR (300 MHz, $CDCl_3$): δ 9.94 (s, 1H), 8.48 (s, 1H), 7.78 (d, *J* = 8.4 Hz, 2H), 7.27 (d, *J* = 8.4 Hz, 2H), 3.23 – 3.26 (m, 2H), 2.41 (s, 3H), 1.16 – 1.51 (m, 11H), 0.91 (t, *J* = 7.5 Hz, 3H)

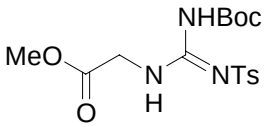
^{13}C NMR (75 MHz, $CDCl_3$): δ 152.73, 150.92, 142.42, 140.64, 129.35, 126.04, 84.34, 43.21, 30.0*, 28.28, 22.42, 21.77,11.61.

HRMS: *m/z* calculated for $C_{16}H_{26}N_3O_4S$ [M+H] = 356.1651; found 356.1644



^{1}H NMR (300 MHz, $CDCl_3$, ppm): δ 9.86 (s, 1H), 8.47 (s, 1H), 7.79 (d, *J* = 8.4 Hz, 2H), 7.27 (d, *J* = 8.4 Hz, 2H), 2.41 (s, 3H), 1.50 (s, 9H), 1.37 (s, 9H).

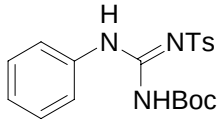
^{13}C NMR (300 MHz, $CDCl_3$, ppm): δ 152.87, 149.30, 142.31, 140.69, 129.36, 125.94, 84.17, 52.88, 30.0*, 28.98, 28.31, 21.79.
HRMS: *m/z* calculated for $C_{17}H_{28}N_3O_4S$ [M+H] = 370.1800; found 370.1801



^{1}H NMR (300 MHz, $CDCl_3$): δ 9.96 (s, 1H), 8.96 (s, 1H), 7.77 (d, *J* = 8.4Hz, 2H), 7.27 (d, *J* = 8.4 Hz, 2H), 4.09 (d, *J* = 5.1 Hz, 2H), 3.70 (s, 3H), 2.41 (s, 3H), 1.52 (s, 9H).

^{13}C NMR (75 MHz, $CDCl_3$): δ 169.17, 152.37, 150.94, 142.74, 140.15, 129.38, 126.13, 84.78, 52.72, 43.12, 30.0*, 28.24, 21.79

HRMS: *m/z* calculated for $C_{16}H_{24}N_3O_6S$ [M+H] = 386.1386; found 386.1392



^{1}H NMR (300 MHz, $CDCl_3$): δ 10.45 (s, 1H), 10.12 (s, 1H), 7.82 (d, *J* = 8.4 Hz, 2H), 7.35 (d, *J* = 8.4 Hz, 2H), 7.33 – 7.28 (m, 4H), 7.15 (t, *J* = 7.5 Hz, 1H), 2.42 (s, 3H), 1.56 (s, 3H).

^{13}C NMR (75 MHz, $CDCl_3$): δ 152.82, 148.19, 142.83, 140.02, 135.95, 129.50, 128.97, 126.18, 125.61, 122.35, 85.10, 30.0*, 28.30, 21.80

HRMS: *m/z* calculated for $C_{19}H_{24}N_3O_4S$ [M+H] = 390.1487; found 390.1485

* Signals due to tBu groups of Boc rotamers.

Electrospray Mass Spec of Peptide Products, Purified by RP-HPLC:

Peptide	MW_{calc}	Sequence
4	1758.0	KKK**X**$_{\mathbf{Gdn}}$AQEKELQAL
5	1899.3	KKK**K**$_{\mathbf{Gdn}}$AQLEKELQAL
6	3688.6	AQLKKKLQAL**X**$_{\mathbf{Gdn}}$KKNAQLKK**X**$_{\mathbf{Gdn}}$LQALKKKLAQ
7	3674.5	AQLKKKLQALKK**X**$_{\mathbf{Gdn}}$NAQL**X**$_{\mathbf{Gdn}}$K**X**$_{\mathbf{Gdn}}$LQAL**X**$_{\mathbf{Gdn}}$KKLAQ
8	3772.4	AQL**X**$_{\mathbf{Gdn}}$K**X**$_{\mathbf{Gdn}}$LQAL**X**$_{\mathbf{Gdn}}$K**X**$_{\mathbf{Gdn}}$NAQL**X**$_{\mathbf{Gdn}}$K**X**$_{\mathbf{Gdn}}$LQAL**X**$_{\mathbf{Gdn}}$K**X**$_{\mathbf{Gdn}}$LAQ