

2-Aminoresorcinol: A solution of 22.4 g (129 mmol) of sodium dithionite in 104 mL of water was added dropwise to a solution of 5 g (32 mmol) of 2-nitroresorcinol in 320 mL of ethanol. The resulting mixture was stirred at room temperature for 30 minutes. The precipitate was filtered and the solution evaporated to dryness. The crude was purified by silica gel column chromatography (AcOEt) to yield 2.82 g (22.5 mmol, 70 %) of product. **¹H-NMR** (D₂O, 200 MHz): δ 7.41 (1H, dd, J_a=2 Hz, J_b=6 Hz, H-5); 6.30 (1H, d, J=1, H-4), 6.29 (1H, d, J=1 Hz, H-6); **¹³C-NMR** (D₂O, 50 MHz): δ 144.7 (C-1, C-3); 137.0 (C-2), 118.8 (CH, C-5), 107.3 (CH, C-6); **ES MS** (positive mode): *m/z* 126.3 (M_{calc} for C₆H₇NO₂: 125.1); **mp**: 152.5 °C

N,N'-bis-*Z*-*N''*-(2-Hydroxyethyl)-guanidine: A solution of 8.4 mL (8.5 g, 14 mmol) of ethanolamine in 17 mL of ACN was added dropwise to a solution of 5 g (14 mmol) of *N,N'*-bis-*Z*-methylisothiourea and 7.8 mL (56 mmol) of triethylamine in 170 mL of ACN. The resulting solution was refluxed under N₂ atmosphere for 22 hours. The crude was evaporated to dryness. The resulting solid was suspended in water and washed with CH₂Cl₂. The organic layer was dried over Na₂SO₄, filtered and evaporated to dryness, and the crude was purified by silica gel column chromatography (CH₂Cl₂) to yield 1.70 g (5.07 mmol, 36 %) of product. **¹H-NMR** (CDCl₃, 200 MHz): δ 11.8 (1H, NH), 7.36 (10H, m, aromatics), 5.18 (2H, s, CH₂, Z), 5.11 (2H, s, CH₂, Z), 3.74 (2H, m, CH₂), 3.59 (2H, m, CH₂); **¹³C-NMR** (CDCl₃, 50 MHz): δ 163.1 (C-guan), 156.9 (COO), 153.6 (COO), 136.4 (Cq, Ph), 134.4 (Cq, Ph), 128.8, 128.6, 128.4, 128.3, 127.9 (CH, Ph), 68.3, 67.1 (CH₂, Z), 62.4 (CH₂), 44.2 (CH₂); **ES MS** (positive mode): *m/z* 372.4 (M_{calc} for C₁₉H₂₁N₃O₅: 371.2)

5-Bromo-*N*¹-*tert*-butoxycarbonylmethyl-uracil (**2**): 800 μL (1.056 g, 5.4 mmol) of *tert*-butyl bromoacetate was added to a suspension of 1.03 g (5.4 mmol) of 5-bromouracil and 738 mg (5.4 mmol) of K₂CO₃ in 20 mL of dry DMF. The resulting mixture was vigorously stirred under argon atmosphere, at room temperature for 21 hours. The reaction crude was filtered. The resulting solution was diluted with AcOEt, washed with water, dried over Na₂SO₄, and evaporated to dryness to yield 1.4 g (4.6 mmol, 85 %) of product. **¹H-NMR** (DMSO-*d*₆, 200 MHz): δ 8.12 (1H, s, H uracil), 4.34 (2H, s, CH₂), 1.39 (9H, s, 3xCH₃); **¹³C-NMR** (DMSO-*d*₆, 50 MHz): δ 167.4 (COO), 160.2 (C-4), 150.8 (C-2), 146.1 (C-6), 95.1 (C-Br), 82.7 (Cq), 49.9 (CH₂), 28.3 (CH₃); **FAB MS** (positive mode, NBA): *m/z* 305.1/307.1 (M_{calc} for C₁₀H₁₃BrN₂O₄: 304.1/306.1)

5-Bromo-*N*¹-(*tert*-butoxycarbonylmethyl)-4-*N*¹-(1,2,4-triazolyl)-uracil (**3**): 6.11 g (88.5 mmol) of 1,2,4-triazole were suspended in 300 mL of dry ACN under argon atmosphere, and the mixture was cooled in an ice bath. 1.9 mL (19.7 mmol) of POCl₃ and 20.4 mL (14.9 g, 147.4 mmol) of dry TEA was added and left to react for 30 min. Then a solution of 3 g (9.8 mmol) of **2** in 70 mL of dry ACN was added dropwise through a cannula. Once the addition was over, the ice bath was removed and the mixture left to react at room temperature for 21 hours. The reaction crude was cooled in an ice bath and treated with 5 mL of 1 M aq. triethylammonium hydrogencarbonate. The ACN was removed under vacuo and AcOEt was added. The organic phase was washed with 1 M aq. triethylammonium hydrogencarbonate and brine, dried over Na₂SO₄, filtered and evaporated to dryness to yield 3.17 g (8.9 mmol, 90 %) of product. **¹H-NMR** (CDCl₃, 200 MHz): δ 9.04 (1H, s, H triazole), 8.26 (1H, s, H triazole), 8.05 (1H, s, H uracil), 4.58 (2H, s, CH₂), 1.37 (9H, s, 3xCH₃); **¹³C-NMR** (CDCl₃, 50 MHz): δ 153.7 (CH triazole), 153.4 (CH triazole), 145.1 (C-6 resorcinol), 51.7 (CH₂), 28.0 (CH₃); **ES MS** (positive mode): *m/z* 356.0/358.0 (M_{calc} for C₁₂H₁₄BrN₅O₃: 355.0/357.0)

5-Bromo-(2,6-dihydroxyphenyl)-*N*¹-(*tert*-butoxycarbonylmethyl)-cytosine (**4**):

A: A solution of 305 mg (1 mmol) of **2** and 39.3 mg (1.5 mmol) of Ph₃P in 4 mL of CCl₄-CH₂Cl₂ (1:1) was refluxed for 3 hours under nitrogen atmosphere. The crude was allowed to cool to room

temperature and 138 mg (1.1 mmol) of 2-aminoresorcinol and 0.32 mL (315 mg, 2 mmol) of DBU was added. The resulting solution was stirred at room temperature under nitrogen atmosphere for 13 hours. The crude was evaporated to dryness and redissolved in CH₂Cl₂, washed with an aqueous solution of citric acid (5 %), dried over Na₂SO₄, filtered and evaporated to dryness. Purification by silica gel column chromatography (AcOEt/ hexanes 1:1) afforded 50 mg (0.12mmol, 12 %) of product.

B. 2.3 mL (2.4 g, 15.5 mmol) of DBU was added to a solution of 2.64 g (7.4 mmol) of **3** and 1.85 g (14.8 mmol) of 2-aminoresorcinol in 50 mL of dry ACN. The resulting solution was stirred at room temperature under argon atmosphere for 20 hours. The crude was evaporated to dryness, redissolved in CH₂Cl₂, and poured into a vigorously stirred aq. citric acid (5 %) solution. After the addition of hexanes, the precipitate formed was filtered off, washed with H₂O, CH₂Cl₂ and ACN to yield 2.46 g (5.95 mmol, 80 %) of product. **¹H-NMR** (CD₃COCD₃, 200 MHz): δ 8.07 (1H, s, H uracil), 6.81 (1H, t, J=8 H-4 resorcinol), 6.35 (2H, d, J=8, H-3, H-5 resorcinol), 4.42 (2H, s, CH₂), 1.33 (9H, s, 3xCH₃); **¹³C-NMR** (CD₃COCD₃, 50 MHz): δ 165.1 (COO), 148.9 (C-2 resorcinol), 146.9 (C-6 resorcinol), 125.8 (C-4 resorcinol), 107.7 (C-3 C-5 resorcinol), 97.1 (C-Br), 81.0 (Cq ^tBu), 49.8 (CH₂), 26.4 (CH₃); **MALDI-TOF MS** (positive mode, 2,5-dihydroxybenzoic acid (DHB)): 412.9/414.9; **MALDI-TOF MS** (negative mode, DHB): *m/z* 410.3/412.3 (M_{calc} for C₁₆H₁₈BrN₃O₅: 411.0/413.0)

*N*¹-(*tert*-Butoxycarbonylmethyl)-6-hydroxyphenoxazine (**5**): 3.2 g (55 mmol) of KF was added to a solution of 2.26 g (5.5 mmol) of **4** in 250 mL of ethanol. The mixture was refluxed under nitrogen atmosphere for 20 hours. The crude was cooled to room temperature, evaporated to dryness and purified by silica gel column chromatography (AcOEt and increasing amounts of MeOH, 0 to 10 %) to yield 1.39 g (4.2 mmol, 76 %) of product. **¹H-NMR** (CD₃COCD₃, 200 MHz): δ 7.15 (s, 1H, H uracil), 6.72 (d, 1H, J = 8 Hz, H-3 resorcinol), 6.53 (dd, 1H, J = 8 Hz, H-4 resorcinol), 6.27 (d, 1H, J = 8 Hz, H-5 resorcinol), 4.38 (s, 2H, CH₂), 1.45 (s, 9H, 3xCH₃); **¹³C-NMR** (CD₃COCD₃, 50 MHz): δ 166.5 (COO), 155.4 (CO), 146.1 (C-6 resorcinol), 129.3 (C-4 resorcinol), 107.9 (C-5 resorcinol), 105.5 (C-3 resorc), 86.8 (Cq, ^tBu), 55.2 (CH₂), 28.0 (CH₃); **MALDI-TOF MS** (positive mode, DHB): 333.9; **MALDI-TOF MS** (negative mode, DHB): *m/z* 329.7 (M_{calc} for C₁₆H₁₇N₃O₅: 331.1)

*N*¹-(*tert*-Butoxycarbonylmethyl)-6-(*N*-Z-2-aminoethoxy)-phenoxazine (**6a**): A suspension of 665 mg (2 mmol) of polymer-bound triphenylphosphine (Fluka), 245 μL (2 mmol) of diethylazodicarboxylate, 480 mg (1.16 mmol) of **5** and 389 mg (2 mmol) of *N*-Z-ethanolamine in 25 mL of dry CH₂Cl₂ was stirred at room temperature under nitrogen atmosphere for 4 hours. The resin was filtered and washed with CH₂Cl₂. The solvent was evaporated and the crude purified by silica gel column chromatography (AcOEt/hexanes 1:1) to yield 271 mg (0.53 mmol, 40 %) of product. **¹H-NMR** (CDCl₃, 200 MHz): δ 11.6 (1H, NH), 10.4 (1H, NH), 7.58 (1H, s, H uracil), 7.36 (5H, m, Z), 6.85 (3H, m, C-H resorcinol), 5.10 (2H, s, CH₂ Z), 4.45 (2H, s, CH₂), 3.82-3.55 (4H, m, 2xCH₂ arm), 1.47 (9H, s, 3xCH₃); **¹³C-NMR** (CDCl₃, 50 MHz): δ 166.3 (COO), 156.3 (COO), 146.4 (C-H uracil), 136.4 (C-H resorcinol), 128.9, 128.8, 128.5, 128.4 (CH Z), 83.6 (Cq ^tBu), 66.7 (CH₂, Z), 62.2 (CH₂ arm), 50.7 (CH₂ arm), 43.5 (CH₂), 28.0 (CH₃); **MALDI-TOF MS** (positive mode, DHB): *m/z* 510.2 (M_{calc} for C₂₆H₂₈N₄O₇: 508.2)

*N*¹-(*tert*-Butoxycarbonylmethyl)-6-[2-(*N,N'*-bis-Z-guanidino)ethoxy]-phenoxazine (**6b**): A suspension of 1 g (3 mmol) of polymer-bound triphenylphosphine (Fluka), 368 μL (3 mmol) of diethylazodicarboxylate, 664 mg (2 mmol) of **5** and 1.11 g (3 mmol) of *N,N'*-bis-Z-*N'*-(2-hydroxyethyl)-guanidine in 25 mL of dry CH₂Cl₂ was stirred at room temperature under nitrogen atmosphere for 4 hours. The resin was filtered and washed with CH₂Cl₂. The solvent was evaporated and the crude purified by silica gel column chromatography (AcOEt/hexanes 1:1) to yield 540 mg (0.79 mmol, 39 %) of product. **¹H-NMR** (CDCl₃, 200 MHz): δ 11.5 (1H, NH), 8.63 (1H, NH), 7.73 (1H, s, H uracil), 7.34 (13H, m, Z+resorcinol), 5.18 (2H, s, CH₂, Z), 5.12 (2H, s, CH₂, Z), 4.37 (2H, s,

CH₂), 3.82-3.40 (4H, m, 2xCH₂ arm), 1.47 (9H, s, 3xCH₃); ¹³C-NMR (CDCl₃, 50 MHz): δ 130.8, 128.7, 128.1 (CH, Z), 65.5 (CH₂ Z), 61.7 (CH₂ arm), 29.7 (CH₃); **MALDI-TOF MS** (positive mode, DHB): *m/z* 682.7 (M_{calc} for C₃₅H₃₆N₆O₉: 684.3)

*N*¹-Carboxymethyl-6-(*N*-Z-2-aminoethoxy)-phenoxazine (**7a**): 2 mL of a 4 M solution of HCl in 1,4-dioxane was added to a solution of 400 mg (0.79 mmol) of **6a** in 4 mL of dry CH₂Cl₂. The reaction mixture was stirred for 16 hours at room temperature. The solvent was partially removed in vacuo and hexanes was added. The solid formed was filtered off, washed with cold CH₂Cl₂, and dried to yield 340 mg (0.75 mmol, 95 %) of product. ¹H-NMR (CDCl₃, 200 MHz): δ 7.34 (6H, m, CH Z, CH uracil), 6.76 (3H, m, CH resorcinol), 5.10 (2H, s, CH₂ Z), 3.68 (2H, m, CH₂ arm), 3.34 (2H, m, CH₂ arm); ¹³C-NMR (CDCl₃, 50 MHz): δ 157.0 (COO), 156.7 (CO), 136.3 (CH resorcinol), 128.4, 128.1, 127.6 (CH, Z), 66.9 (CH₂ Z), 62.2 (CH₂ arm), 43.5 (CH₂); **MALDI-TOF MS** (positive mode, DHB): *m/z* 453.5 (M_{calc} for C₂₂H₂₀N₄O₇: 452.1)

*N*¹-Carboxymethyl-6-[2-(*N,N'*-bis-Z-guanidino)ethoxy]-phenoxazine (**7b**): 8 mL of a 4 M solution of HCl in 1,4-dioxane was added to a solution of 590 mg (0.86 mmol) of **6b** in 15 mL of dry CH₂Cl₂. The reaction mixture was stirred for 17 hours at room temperature. The solvent was partially removed in vacuo and hexanes was added. The solid formed was filtered off, washed with cold CH₂Cl₂ and dried to yield 530 mg (0.84 mmol, 98 %) of product. ¹H-NMR (DMSO-d₆, 200 MHz): δ 11.0 (1H, NH), 8.79 (1H, NH), 7.86 (1H, s, H uracil), 7.37 (13H, sc, CH Z, CH resorcinol), 5.23 (2H, s, CH₂ Z), 5.16 (2H, s, CH₂ Z), 4.09-3.88 (4H, m, 2xCH₂ arm), 3.66 (2H, s, CH₂); ¹³C-NMR (DMSO-d₆, 50 MHz): δ 130.8, 128.7, 128.1 (CH, Z), 65.5 (CH₂ Z), 61.7 (CH₂ Z); **MALDI-TOF MS** (positive mode, DHB): *m/z* 626.3 (M_{calc} for C₃₁H₂₈N₆O₉: 628.2)

tert-Butyl *N*-[2-(*N*-9-fluorenylmethoxycarbonyl)aminoethyl]-*N*-[carboxymethyl-6-(*N*-Z-2-aminoethoxy)-phenoxazine]-glycinate (**8a**): A solution of 400 mg (1.2 mmol) of *tert*-butyl *N*-[2-(*N*-9-fluorenylmethoxycarbonyl)aminoethyl]-glycinate in 10 mL of dry DMF was cooled to 0 °C and 340 mg (0.75 mmol) of **7a** and 288 mg (1.5 mmol) of 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (EDC) was added. The solution was stirred at room temperature for 17 hours. The resulting mixture was poured into ice-water with vigorous stirring. The aqueous layer was washed with CH₂Cl₂. The organic layer was dried over Na₂SO₄, filtered and evaporated to yield 390 mg (0.47 mmol, 63 %) of product. ¹H-NMR (CDCl₃, 200 MHz): δ 7.73 (2H, m, CH Fmoc), 7.61 (2H, d, J=7.5 Hz, CH Fmoc), 7.40-7.26 (13H, m, CH Z, CH Fmoc, CH uracil, CH resorcinol), 5.10 (2H, s, CH₂ Z), 4.39 (2H, d, J=7Hz, CH₂ Fmoc), 4.21 (1H, t, J=7 Hz, CH Fmoc), 3.96 (1H, t, J=6 Hz, H-1 aminoethylglycine), 3.69 (2H, m, CH₂ arm), 3.34 (2H, m, CH₂ arm), 3.29 (2H, s, H-2 aminoethylglycine), 2.74 (2H, t, J=5 Hz, H-3 aminoethylglycine), 1.47 (s, 9H, 3xCH₃); ¹³C-NMR (CDCl₃, 50 MHz): δ 162.4 (COO), 156.8 (COO), 128.6 (CH, Z), 126.9 (CH, Z), 120.8 (CH, Fmoc), 119.6 (CH, Fmoc), 107.7 (CH, resorcinol), 83.0 (CH₂ Fmoc), 55.4 (CH₂, aminoethylglycine), 42.9 (CH₂), 27.9 (CH₃); **MALDI-TOF MS** (positive mode, DHB): *m/z* 831.2 (M_{calc} for C₄₅H₄₆N₆O₁₀: 830.3).

N-[2-(*N*-9-fluorenylmethoxycarbonyl)aminoethyl]-*N*-[carboxymethyl-6-(*N*-Z-2-aminoethoxy)-phenoxazine]glycine (**9a**): 5 mL of TFA 100% was added dropwise to a 1 mL dry CH₂Cl₂ solution of 530 mg (0.64 mmol) of **8a**. The resulting solution was stirred for 30 minutes at 0°C and 30 minutes at room temperature under nitrogen. The reaction mixture was added dropwise to anhyd Et₂O with stirring. The resulting solid was collected by filtration and dried to yield 220 mg (0.28 mmol, 45%) of product. ¹H-NMR (DMSO-d₆, 200 MHz): δ 7.74 (2H, m, CH Fmoc), 7.62 (2H, d, 7.5Hz, CH Fmoc), 7.40-7.25 (13H, m, CH Z, CH Fmoc, CH uracil, CH Resorcinol), 5.09 (2H, s, CH₂ Z), 4.37 (2H, d, 7Hz, CH₂ Fmoc), 4.22 (1H, t, 7Hz, CH Fmoc), 3.97 (1H, t, 6Hz, H-1 aminoethylglycine), 3.69 (2H, m,

CH₂ arm), 3.32 (2H, m, CH₂ arm), 3.28 (2H, s, H-2 aminoethylglycine), 2.73 (2H, t, 5Hz, H-3 aminoethylglycine); ¹³C-NMR (DMSO-d₆, 50 MHz): δ 162.4 (COO), 156.8 (COO), 128.6 (CH, Z), 126.9 (CH, Z), 120.8 (CH, Fmoc), 119.6 (CH, Fmoc), 107.7 (CH, resorcinol), 83.0 (CH₂ Fmoc), 55.4 (CH₂, aminoethylglycine), 42.9 (CH₂, aminoethylglycine); **MALDI-TOF-MS** (positive mode, DHB): *m/z*: 774.6 (M_{calc} for C₄₁H₃₈N₆O₁₀: 774.3)

tert-Butyl *N*-[2-(*N*-9-fluorenylmethoxycarbonyl)aminoethyl]-*N*-[carboxymethyl-6-[2-(*N,N'*-bis-*Z*-guanidinoethoxy)]-phenoxazine]glycinate (**8b**): A dry DMF solution of 303 mg (0.76 mmol) of *tert*-butyl *N*-[2-(*N*-9-fluorenylmethoxycarbonyl)aminoethyl] glycinate was cooled to 0°C and 530 mg (0.84 mmol) of **7b** and 322 mg (1.68 mmol) of 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (EDC) was added. The solution was stirred at room temperature for 5 days. The resulting mixture poured into ice-water with vigorous stirring. The precipitate was collected by filtration to yield 600 mg (0.60 mmol, 71%) of product. ¹H-NMR (CDCl₃, 200 MHz): δ 7.74 (2H, m, CH Fmoc), 7.61 (2H, d, 7Hz, CH, Fmoc), 7.38-7.26 (17H, m, CH Z, CH Fmoc, CH uracil, CH resorcinol), 5.26 (2H, s, CH₂ Z), 5.15 (2H, s, CH₂ Z), 4.36 (2H, m, CH₂ Fmoc), 4.23 (1H, t, 7Hz, CH Fmoc), 3.89 (2H, m, H-1 aminoethylglycine), 3.66 (4H, m, 2xCH₂ arm), 3.27 (2H, m, H-2 aminoethylglycine), 2.75 (2H, m, H-3 aminoethylglycine), 1.47 (s, 9H, 3xCH₃); ¹³C-NMR (CDCl₃, 50 MHz): δ 162.5 (COO), 156.9 (COO), 143.9 (CH, uracil), 141.2 (CH, Fmoc), 140.0 (CH, Fmoc), 137.9 (CH, Fmoc), 136.4 (CH Fmoc), 128.6 (CH, Z), 128.4 (CH, Z), 128.0 (CH, Z), 127.6 (CH, Z), 126.9 (CH, Z), 125.0 (CH, Fmoc), 120.9 (CH, Fmoc), 119.8 (CH, Fmoc), 107.7 (CH, resorcinol), 97.1 (Cq, uracil), 81.5 (CH₂ Fmoc), 66.7 (CH₂ Z), 47.2 (CH₂, aminoethylglycine), 28.1 (CH₃); **MALDI-TOF-MS** (positive mode, DHB): *m/z*: 1005.1 (M_{calc} for C₅₄H₅₄N₈O₁₂: 1006.4)

N-[2-(*N*-9-fluorenylmethoxycarbonyl)aminoethyl]-*N*-[carboxymethyl-6-[2-(*N,N'*-bis-*Z*-guanidinoethoxy)]-phenoxazine]glycine (**9b**): 4 mL of TFA 100% was added dropwise to a 1 mL dry CH₂Cl₂ solution of 146 mg (0.15 mmol) of **8b**. The resulting solution was stirred for 30 minutes at 0°C and 1 hour at room temperature under nitrogen. The resulting mixture was concentrated to one fifth volume in vacuo and the resulting crude was added dropwise to anhyd Et₂O with stirring. The resulting solid was collected by filtration to yield 114 mg (0.12 mmol, 83%) of product. ¹H-NMR (DMSO-d₆, 300 MHz): δ 7.92 (2H, m, Fmoc), 7.69 (2H, d, 7Hz, Fmoc), 7.38-7.25 (14H, m, CH Z, CH Fmoc, CH uracil), 6.89 (1H, m, resorcinol), 6.59 (1H, m, resorcinol), 6.47 (1H, m, resorcinol), 5.14 (2H, s, CH₂ Z), 5.10 (2H, s, CH₂ Z), 4.37 (2H, m, CH₂ Fmoc), 4.24 (1H, m, CH Fmoc), 3.91 (2H, s, CH₂), 3.04 (2H, m, H-2 aminoethylglycine), 2.78 (2H, m, H-3 aminoethylglycine); ¹³C-NMR (DMSO-d₆, 75 MHz): δ 168.8 (COO), 144.5 (CH, uracil), 141.5 (CH, Fmoc), 128.4 (CH, Z), 127.8 (CH, Z), 127.3 (CH, Z), 125.8 (CH, Fmoc), 120.8 (CH, Fmoc), 117.6 (CH, Fmoc), 102.2 (CH, resorcinol), 81.3 (CH₂ Fmoc), 75.6 (CH, Fmoc), 68.4 (CH₂, aminoethylglycine), 66.3 (CH₂, aminoethylglycine), 47.4 (CH₂, aminoethylglycine); **MALDI-TOF-MS** (positive mode, DHB): *m/z*: 948.9 (M_{calc} for C₅₀H₄₆N₈O₁₂: 950.3).