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Supporting Information

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Specific Fluorescent Probe for 8-Oxoguanosine

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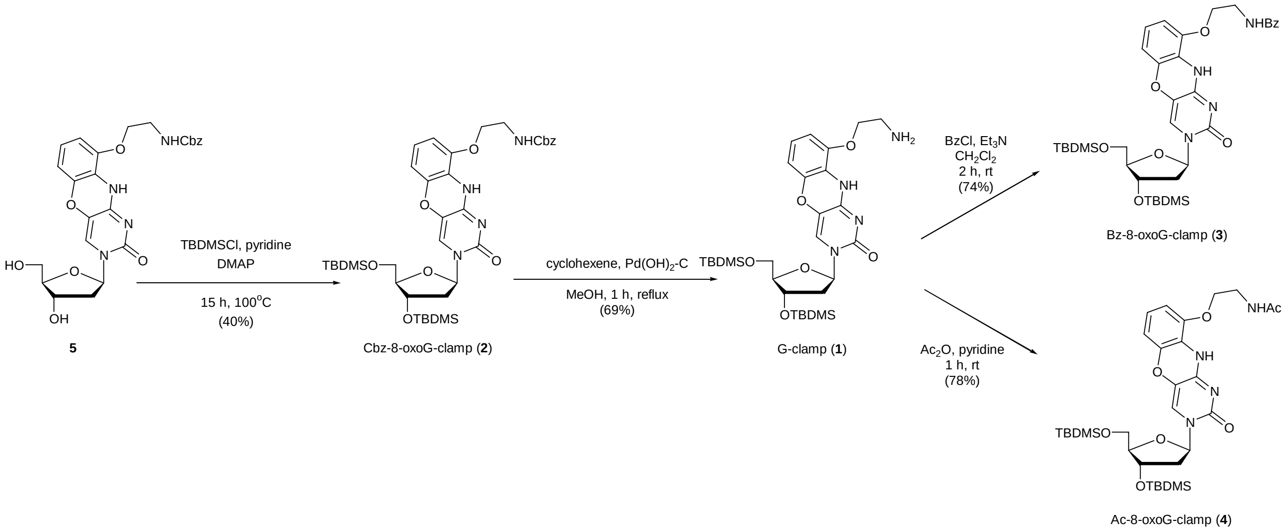
Experimental Procedures



Scheme S1. Synthesis of 8-oxoG-clamps.

Cbz-8-oxoG-clamp (2). Under Ar atmosphere *t*-butyldimethylsilane chloride (336 mg, 2.23 mmol) and 4-(dimethylamino)pyridine (9 mg, 0.07 mmol) were added to a solution of **5**[1] (380 mg, 0.74 mmol) in anhydrous pyridine (20 ml), the mixture was stirred for 15 h at 100 ºC. After the addition of saturated $NaHCO_3$ solution, the mixture was extracted with AcOEt, washed with water and brine, and dried over Na_2SO_4. The solvents were removed under reduced pressure and the residue was purified by flash column chromatography ($CHCl_3$/MeOH 98:2) afforded **2** (222 mg, 40%) as yellow crystals. mp 83-86ºC. $[\alpha]_D^{26}$ -11.5 (*c* 1.00, $CHCl_3$). IR ν_{max} (film): 2930, 1677, 1559, 1498, 1474, 1256, 1098 cm^{-1}. ^{1}H-NMR (400 MHz, $CDCl_3$) δ: 0.05 (6H, s), 0.13 (3H, s), 0.15 (3H, s), 0.86 (9H, s), 0.94 (9H, s), 2.02-2.10 (1H, m), 2.33-2.40 (1H, m), 3.57 (2H, brd, *J* = 5 Hz), 3.76 (1H, d, *J* = 11 Hz), 3.87-3.89 (1H, m), 3.92 (1H, d, *J* = 11 Hz), 4.05 (2H, t, *J* = 4 Hz), 4.37 (1H, dt, *J* = 5, 6 Hz), 5.12 (2H, s), 5.53 (1H, brs), 6.24 (1H, t, *J* = 6 Hz), 6.37 (1H, d, *J* = 8 Hz), 6.45 (1H, d, *J* = 8 Hz), 6.77 (1H, t, *J* = 8 Hz), 7.27-7.38 (5H, m), 7.62 (1H, s). High-resolution MS (ESI): Calcd for $C_{37}H_{55}N_4O_8Si_2$ $(M+H)^+$: 739.3553. Found: 739.3537.
1) K.-Y. Lin, M. D. Matteucci, *J. Am. Chem. Soc.* **1998**, *120*, 8531-8532.

G-clamp (1). Under Ar atmosphere 20% $Pd(OH)_2$-C (98 mg) and cyclohexene (0.36 ml) were added to a solution of **2** (87 mg, 0.12 mmol) in MeOH (3 ml) at room temperature and the mixture was refluxed for 1 h. The reaction mixture was filtered to remove the catalyst, and the solvent was concentrated under reduced pressure. The residue was purified by flash column chromatography ($CHCl_3$/MeOH 95:5, 1% Et_3N) afforded **1** (49 mg, 69%) as yellow crystals. mp 130-133ºC. $[\alpha]_D^{25}$ -15.9 (*c* 1.47, $CHCl_3$). IR ν_{max} (film): 2928, 1670, 1558, 1499, 1474, 1257, 1098 cm^{-1}. ^{1}H-NMR (400 MHz, $CDCl_3$) δ: 0.05 (6H, s), 0.10 (3H, s), 0.12 (3H, s), 0.87 (9H, s), 0.93 (9H, s), 1.90-1.94 (1H, m), 2.22-2.28 (1H, m), 3.47-3.58 (2H, m), 3.72 (1H, d, *J* = 11 Hz), 3.83 (1H, m), 3.87 (1H, d, *J* = 11 Hz), 4.19 (1H, m), 4.31 (2H, brd, *J* = 5 Hz), 6.05 (1H, t, *J* = 4 Hz), 6.21 (1H, d, *J* = 8 Hz), 6.36 (1H, d, *J* = 8 Hz), 6.64 (1H, t, *J* = 8 Hz), 7.31 (1H, brs). High-resolution MS (ESI): Calcd for $C_{29}H_{49}N_4O_6Si_2$ $(M+H)^+$: 605.3185. Found: 605.3191.

Bz-8-oxoG-clamp (3). Under Ar atmosphere benzoyl chloride (3 μl, 0.03 mmol) and Et_3N (7 μl, 0.05 mmol) were added to a solution of **1** (15 mg, 0.025 mmol) in anhydrous dichloromethane (2 ml) at 0 ºC and the mixture was stirred at room temperature for 2 h. The reaction mixture was quenched by the addition of saturated $NaHCO_3$ solution. The mixture was extracted with CH_2Cl_2. The organic phase was washed with water and brine and dried over Na_2SO_4 and concentrated under reduced pressure. The residue was purified by silica gel column chromatography (AcOEt) to give **3** (13 mg, 74%) as yellow oil. mp 89-90ºC. $[\alpha]_D^{25}$ 19.5 (*c* 1.05, $CHCl_3$). IR ν_{max} (film): 2929, 1718, 1661, 1564, 1474, 1257, 1090 cm^{-1}. ^{1}H-NMR (400 MHz, $CDCl_3$) δ: 0.04 (3H, s), 0.04 (3H, s), 0.12 (3H, s), 0.14 (3H, s), 0.86 (9H, s), 0.93 (9H, s), 2.00-2.07 (1H, m), 2.30-2.37 (1H, m), 3.75 (1H, d, *J* = 10 Hz), 3.88-3.94 (4H, m), 4.14 (2H, t, *J* = 5 Hz), 4.34-4.38 (1H, m), 6.19 (1H, t, *J* = 6 Hz), 6.34 (1H, d, *J* = 8 Hz), 6.47 (1H, d, *J* = 8 Hz), 6.85 (1H, t, *J* = 9 Hz), 7.38-7.50 (3H, m), 7.72 (1H, s), 7.91 (1H, brs), 7.95 (2H, d, *J* = 7 Hz). High-resolution MS (ESI): Calcd for $C_{36}H_{53}N_4O_7Si_2$ $(M+H)^+$: 709.3447. Found: 709.3391.

Ac-8-oxoG-clamp (4). Under Ar atmosphere acetic anhydride (6 μl, 0.07 mmol) was added to a solution of **1** (30 mg, 0.065 mmol) in anhydrous pyridine (0.500 ml), the mixture was stirred for 1 h at room temperature. After the addition of saturated $NaHCO_3$ solution, the organic phase was extracted with AcOEt, washed with water and brine, and dried over Na_2SO_4. The solvent was removed under reduced pressure and the residue was purified by silica gel column chromatography ($CHCl_3$/MeOH 19:1) afforded **4** (33 mg, 78%) as yellow crystals. mp 88-92ºC. $[\alpha]_D^{28}$ -9.07 (*c* 0.90, $CHCl_3$). IR ν_{max} (film): 3282, 2928, 1671, 1557, 1498, 1473, 1256, 1086 cm^{-1}. ^{1}H-NMR (400 MHz, $CDCl_3$) δ: 0.04 (6H, s), 0.12 (3H, s), 0.14 (3H, s), 0.86 (9H, s), 0.94 (9H, s), 1.99 (3H, s), 2.02-2.09 (1H, m), 2.32-2.35 (1H, m), 3.63 (2H, q, *J* = 5, 10 Hz), 3.75 (1H, d, *J* = 10 Hz), 3.87 (1H, m), 3.91 (1H, d, *J* = 10 Hz), 4.04 (2H, t, *J* = 4 Hz), 4.34-4.39 (1H, m), 6.23 (1H, t, *J* = 6 Hz), 6.33 (1H, t, *J* = 8 Hz), 6.42 (1H, d, *J* = 8 Hz), 6.75 (1H, d, *J* = 8 Hz), 7.59 (1H, s). High-resolution MS (ESI): Calcd for $C_{31}H_{51}N_4O_7Si_2$ $(M+H)^+$: 647.3158. Found: 647.3291.

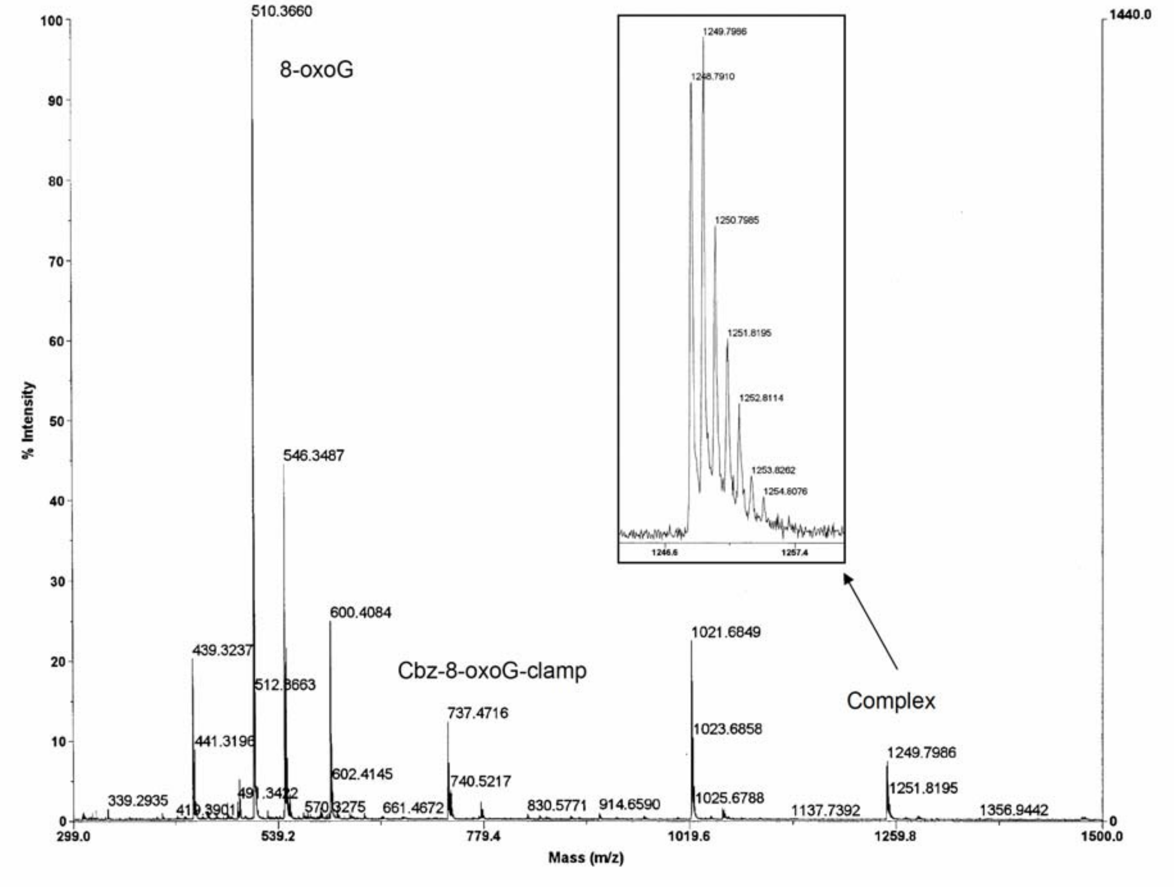


Figure S1. Analysis of the complex of Cbz-8-oxoG-clamp (**2**):8-oxo-dG by negative ESI-MS measurement. Cbz-8-oxoG-clamp (**2**): 737.47 $(M-H)^-$ (Calcd. 737.34), 8-oxo-dG: 510.37 $(M-H)^-$ (Calcd. 510.26), complex: 1248.8 $(M-H)^-$ (Calcd. 1248.6).

Table S1. Binding constants of G-clamp (**1**) and 8-oxoG-clamps (**2-4**) toward 8-oxo-dG and dG in $CHCl_3$.[a]

G-clamp (X=)	K_s (M^{-1})				
	8-oxo-dG	dG	dA	dC	dT
1 (H)	2.7×10^6	1.8×10^7	nq	nq	nq
2 (Cbz)	7.1×10^6	6.9×10^5	nq	nq	nq
3 (Bz)	2.5×10^6	3.9×10^6	nq	nq	nq
4 (Ac)	1.3×10^6	4.0×10^6	nq	nq	nq

[a] Conditions: $CHCl_3$, 0-10 μM 8-oxo-dG or dG, 1 μM G-clamp (**1**) or 8-oxoG-clamps (**2-4**) at 25ºC, excitation 365 nm. **1-4** and target nucleosides (8-oxo-dG, dG, dA, dC and dT): 3'*O*,5'*O*-di-*tert*-butyldimethylsilyl- 2'-deoxy-nucleoside. nq: no fluorescence quenching.

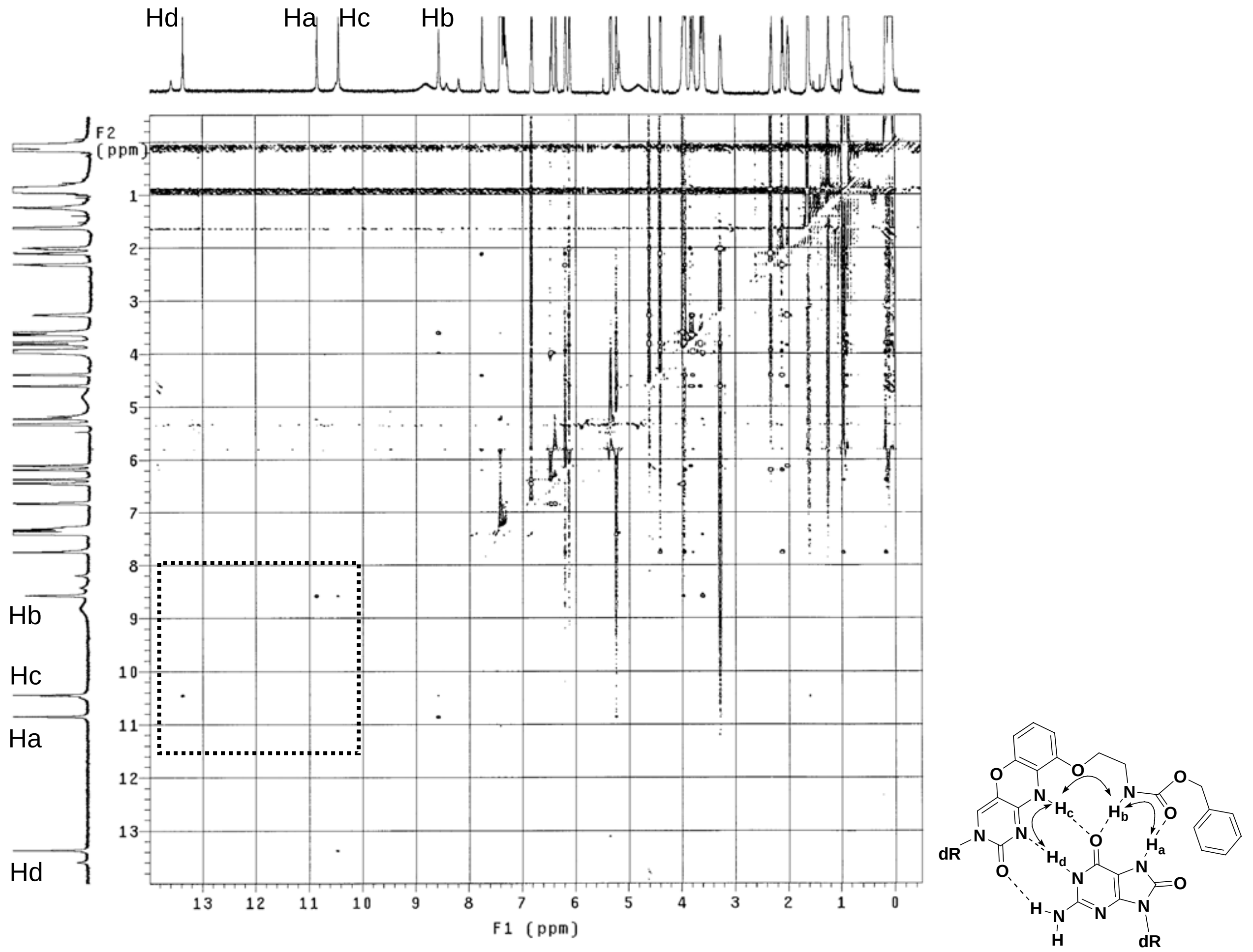


Figure S2. ROESY spectrum of a 1:1 mixture of Cbz-8-oxoG-clamp (**2**) and 8-oxo-dG in CD_2Cl_2 at -10ºC (mixing time = 300 ms). dR = 3'*O*,5'*O*-di-*tert*-butyldimethylsilyl-β-D-2'-deoxyribofuranosyl. The area indicated by a dotted square is shown in Figure 4(d).

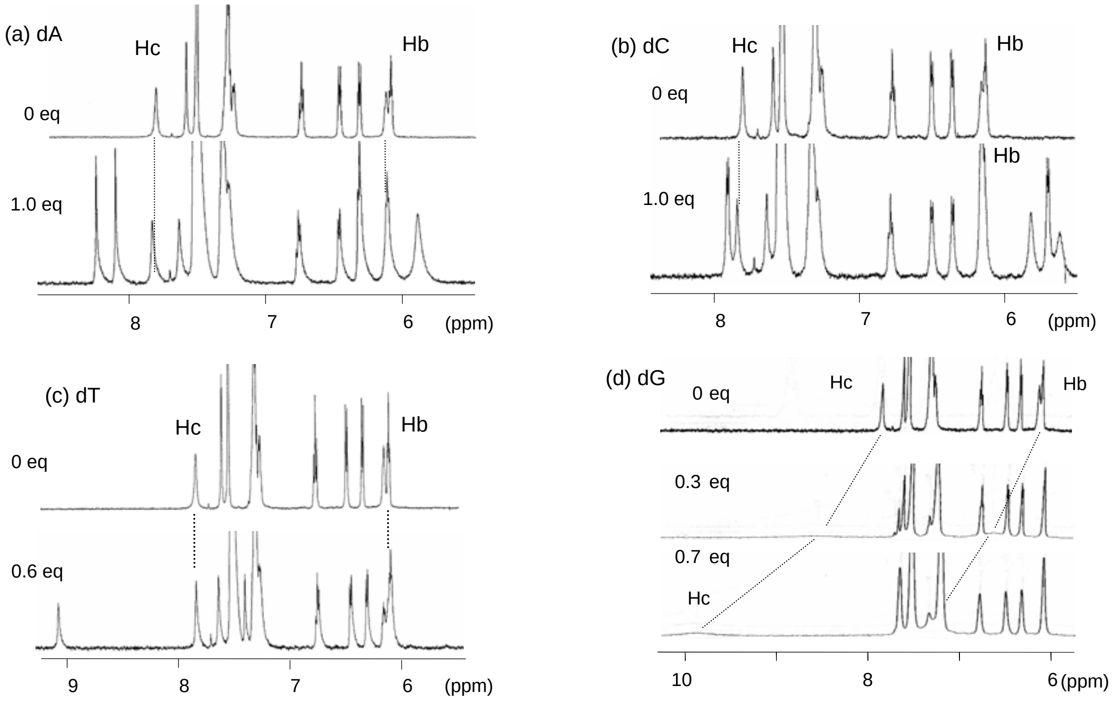


Figure S3. ^{1}H-NMR Change by titration. 3'*O*,5'*O*-di-*tert*-butyldimethylsilyl derivative of dA(a), dC(b) dT(c) or dG (d) was added to Cbz-8-oxoG-clamp (**2**) (2 mM) in $CDCl_3$-CD_3CN (1 : 6) at -10 ºC.

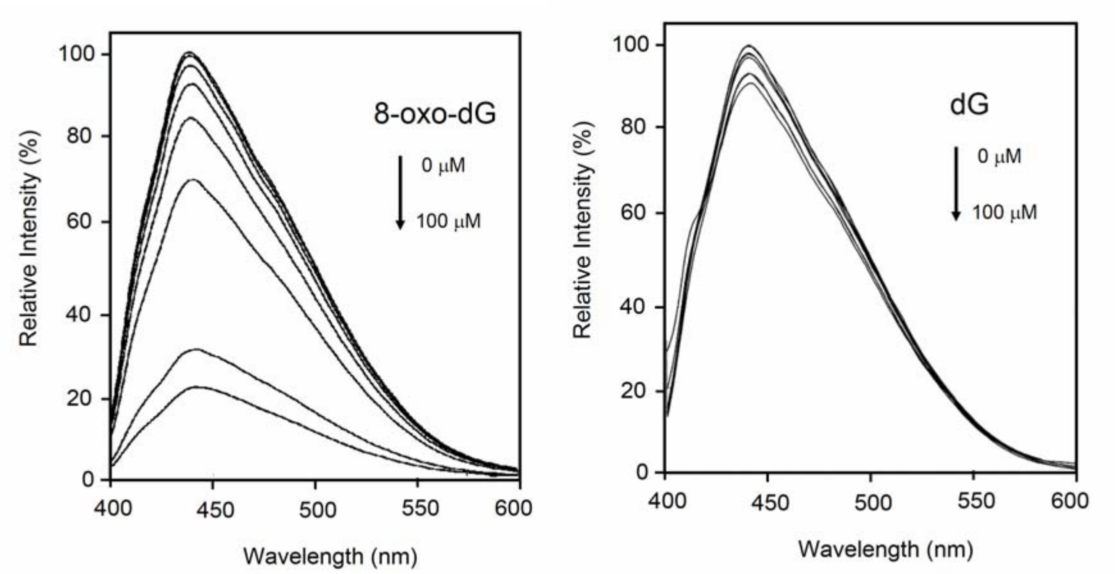


Figure S4. Fluorescence titration spectra of Cbz-8-oxoG-clamp (**2**) in aqueous media. Conditions: 0-100 μM target nucleosides, 1 μM Cbz-8-oxoG-clamp in water buffered with 2.5 mM Triton X-100 at 25ºC, excitation 365 nm. **2** and target nucleosides (8-oxo-dG, dG): 3'*O*,5'*O*-di-*tert*- butyldimethylsilyl-2'-deoxy-nucleoside.

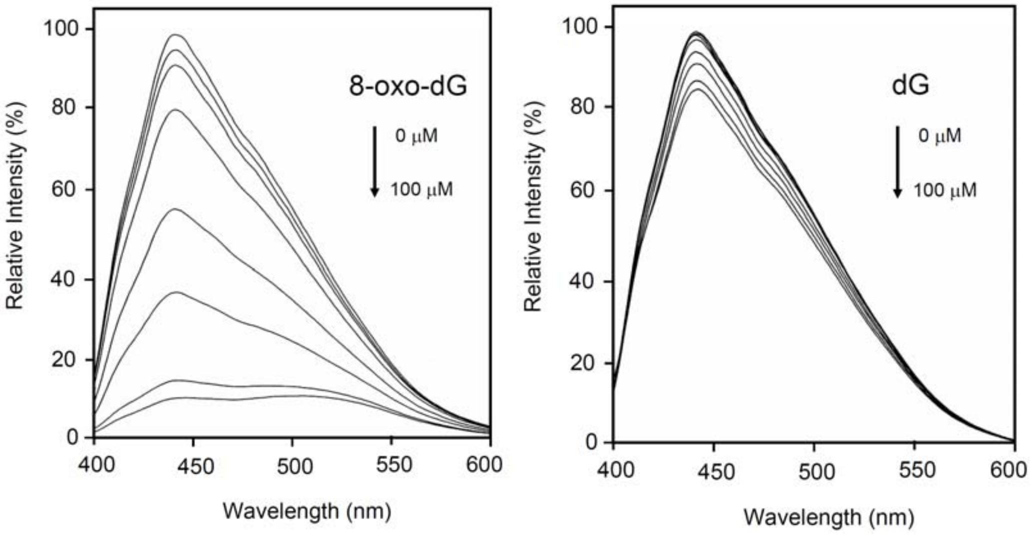


Figure S5. Fluorescence titration spectra of Cbz-8-oxoG-clamp (**2**) with dansylamide in aqueous media. Conditions: 0-100 μM target nucleosides, 1 μM Cbz-8-oxoG-clamp, 2 μM dansylamide in water buffered with 2.5 mM Triton X-100 at 25ºC, excitation 365 nm. **2** and target nucleosides (8-oxo-dG, dG): 3'*O*,5'*O*-di-*tert*-butyldimethylsilyl-2'-deoxy-nucleoside.