Oxovanadium(IV) complexes of phenanthroline bases: the dipyridophenazine complex as a near-IR photocytotoxic agent

Pijus K. Sasmal,[a] Sounik Saha, [a] Ritankar Majumdar,[b] Swati De,[a] Rajan R. Dighe, [b] and Akhil R. Chakravarty*[a]

[a] *Department of Inorganic and Physical Chemistry, Indian Institute of Science, Bangalore 560012, India. Fax: (+91)80-23600683; Tel: +91-80-22932533; E-mail: arc@ipc.iisc.ernet.in*

[b] *Department of Molecular Reproduction, Development and Genetics, Indian Institute of Science, Bangalore 560012, India.*

Electronic Supplementary Information

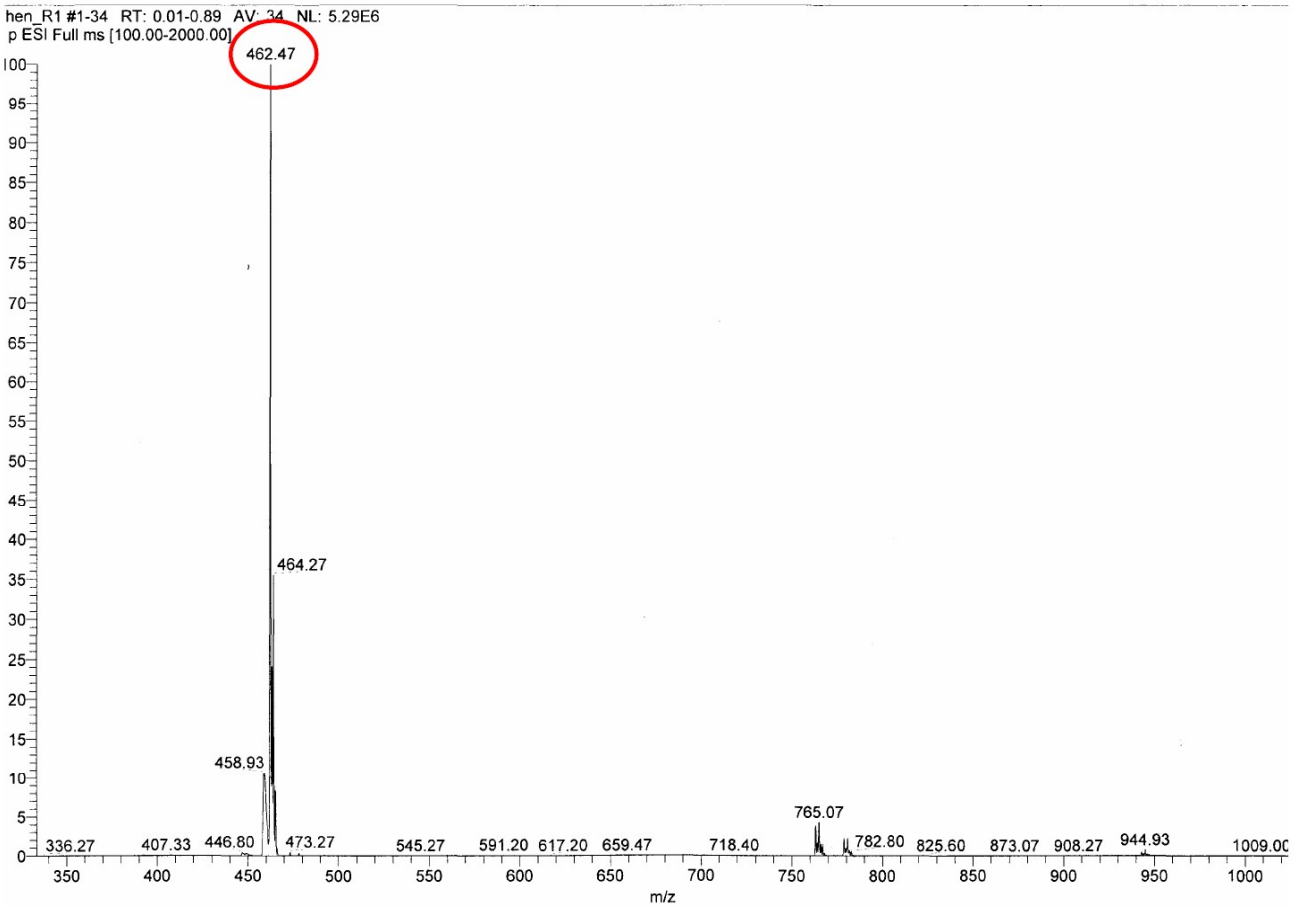

Fig. S1. The ESI-MS spectrum of complex **1** showing the parent ion peak at 462.47 (m/z) in MeOH**.**

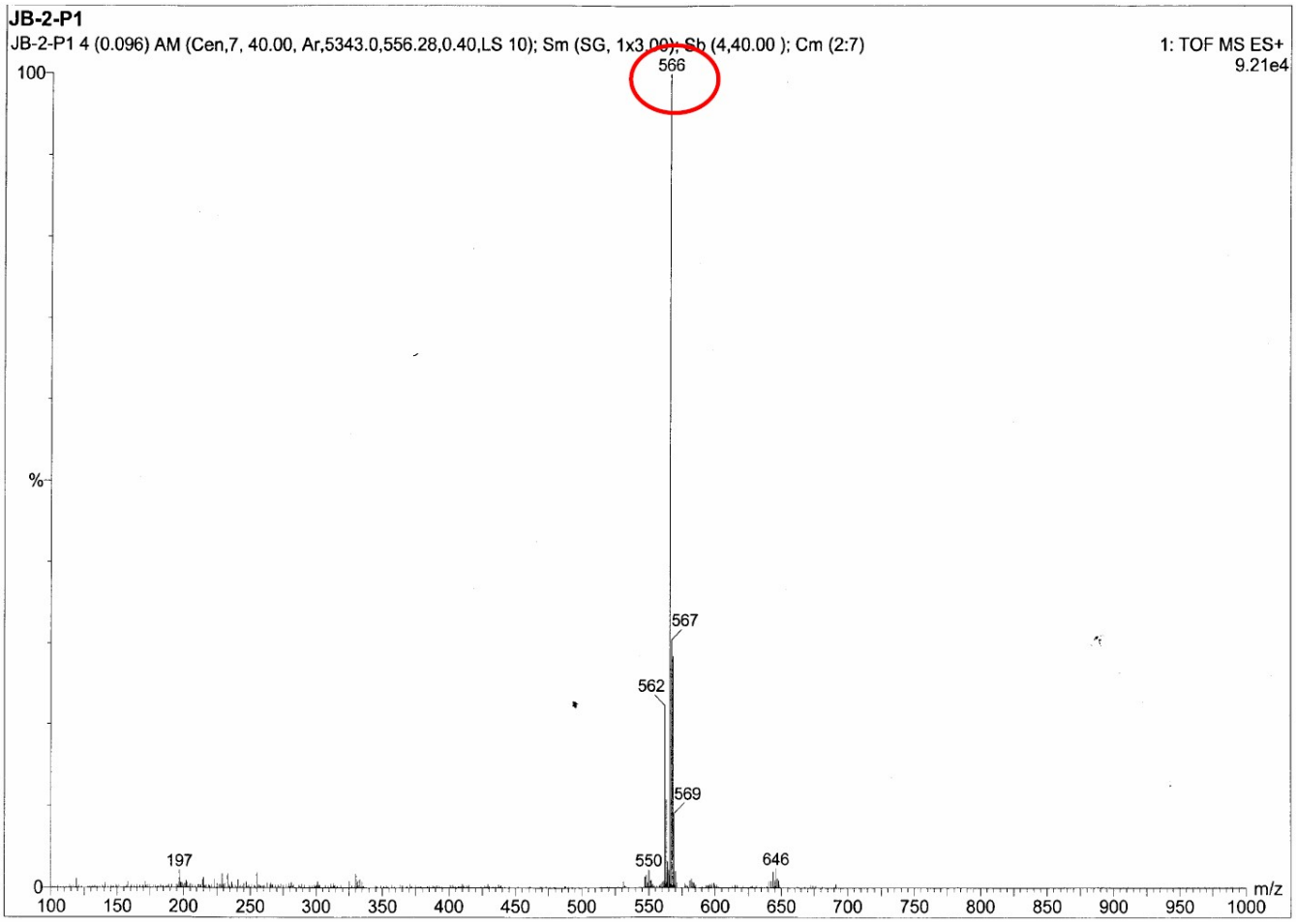

Fig. S2. The ESI-MS spectrum of complex **2** showing the parent ion peak at 566 (m/z) in MeOH**.**

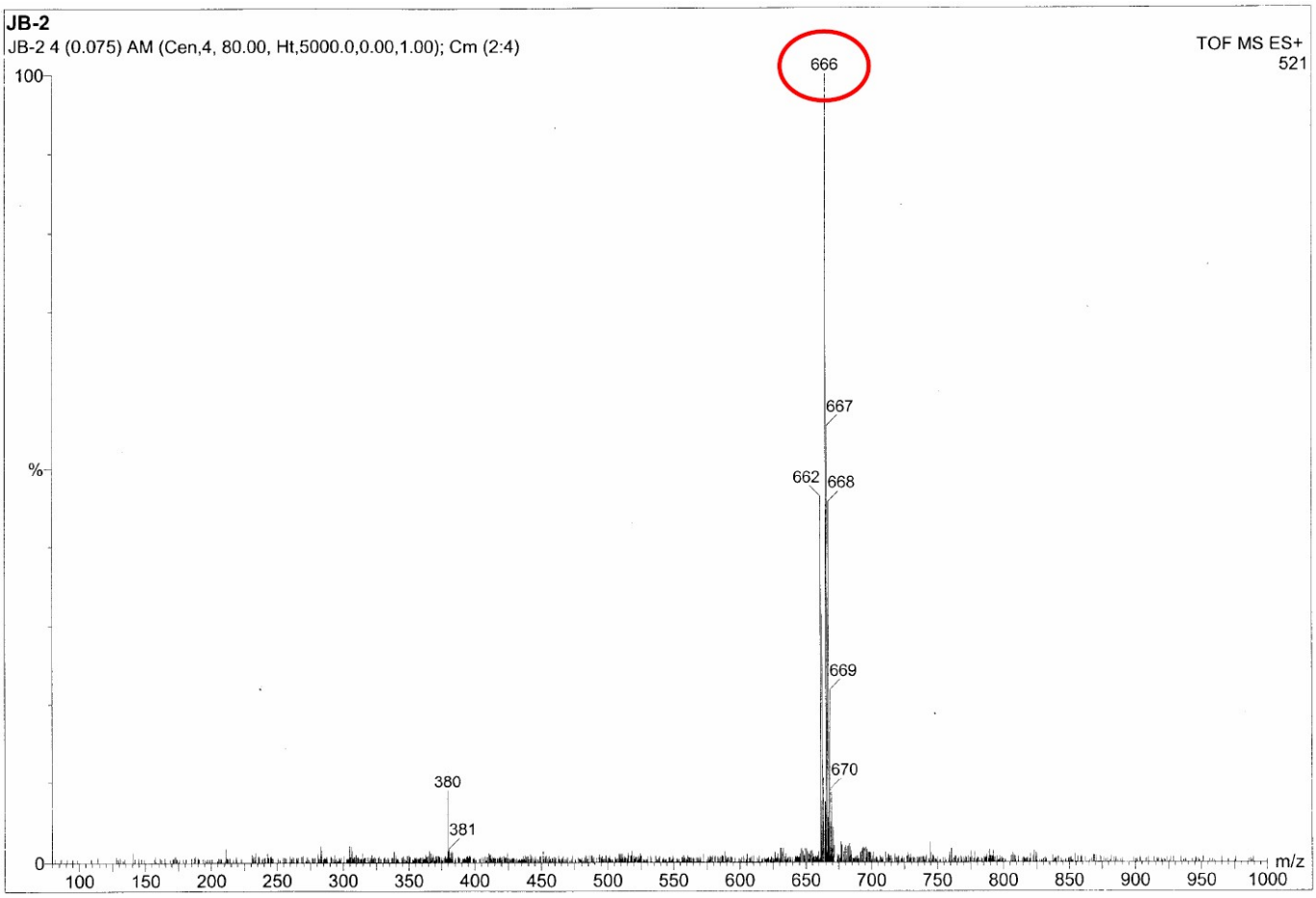

Fig. S3. The ESI-MS spectrum of complex **3** showing the parent ion peak at 666 (m/z) in MeOH**.**

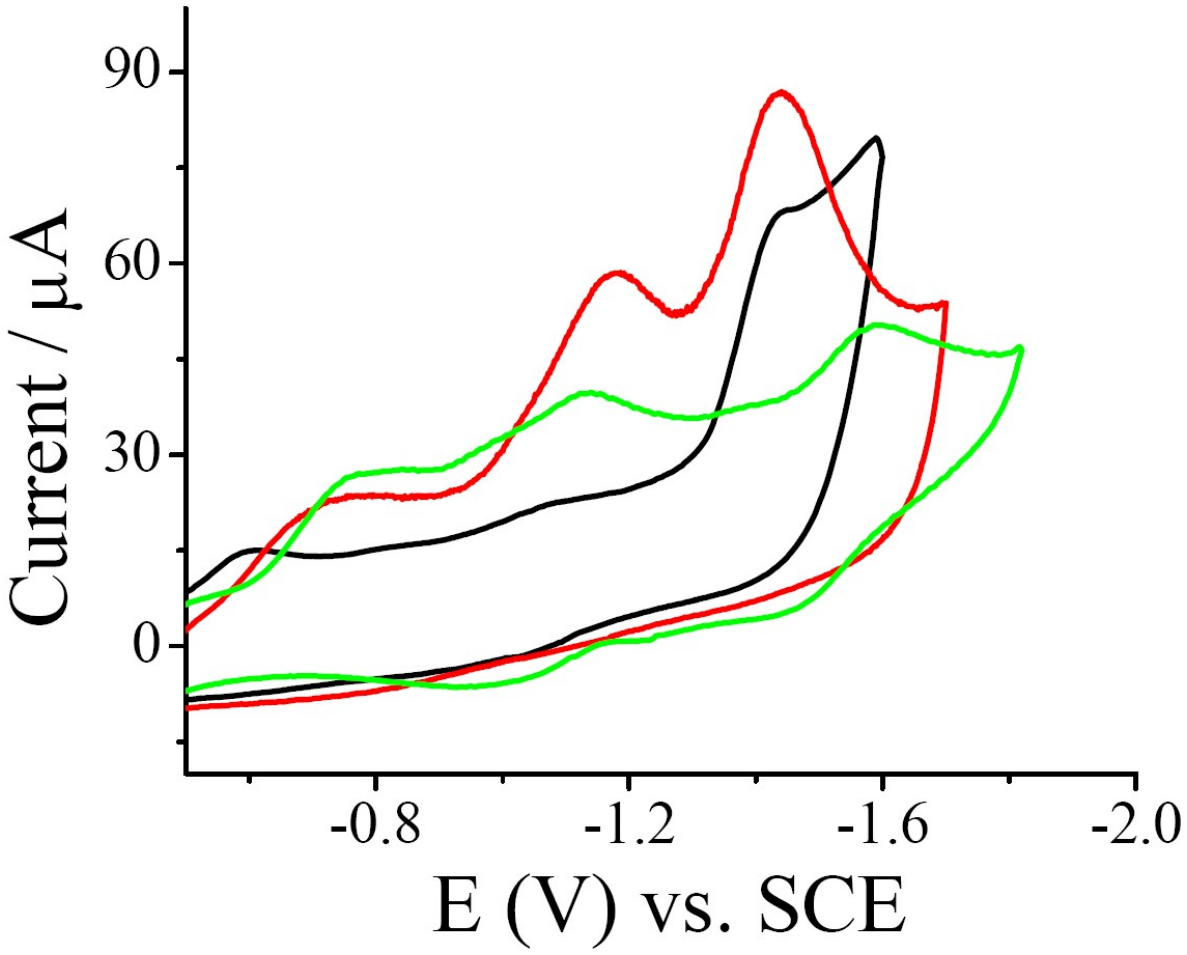

Fig. S4. Cyclic voltammograms of the complexes (**1**) (black), (**2**) (red) and **3** (green) in 20% DMF-Tris-HCl buffer at a scan rate of 50 mV s^{-1} using 0.1 mol KCl as the supporting electrolyte.

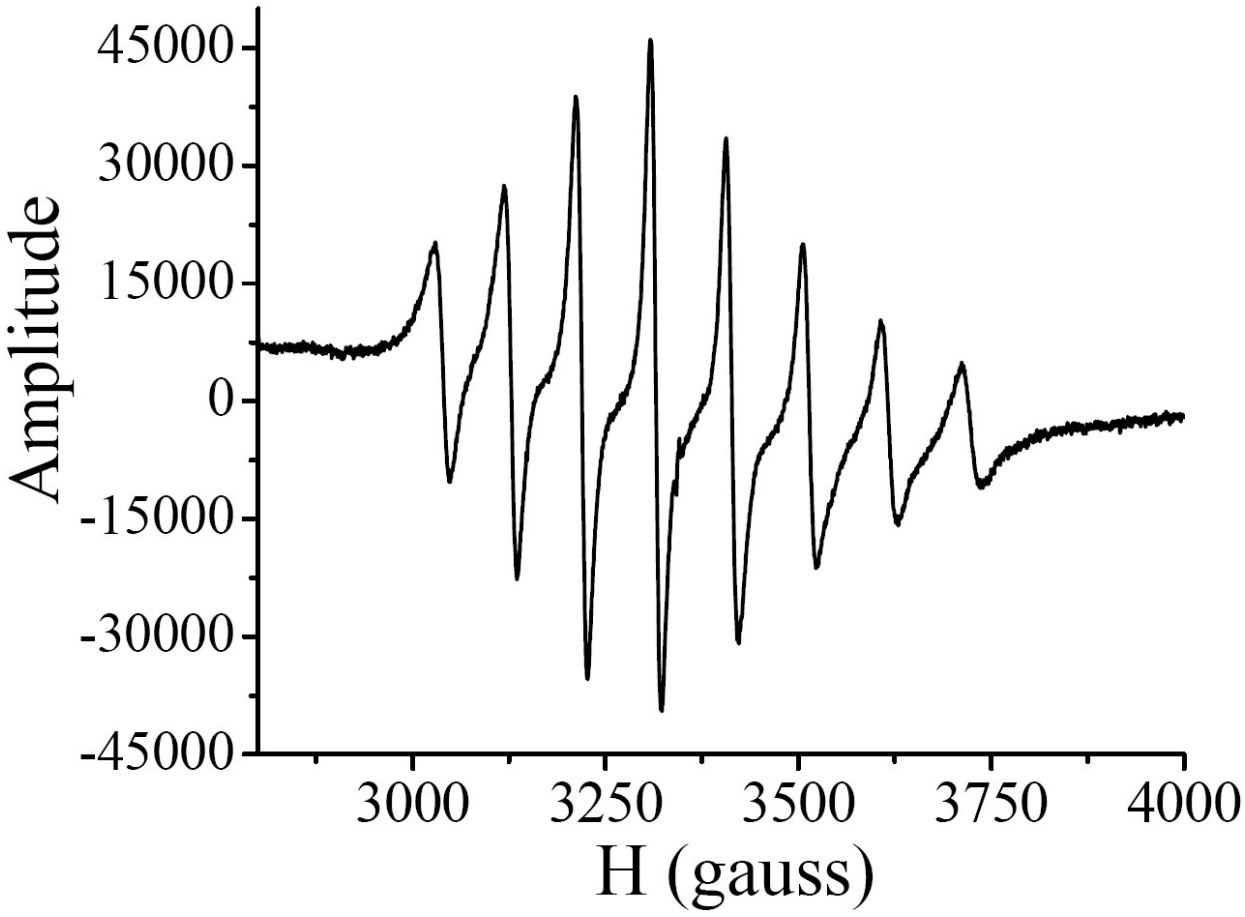

Fig. S5. EPR spectrum of complex **3** (5 mM) in MeOH. The experimental conditions and operating frequency are T = 298 K, ν = 9.39 GHz, modulation amplitude = 4.0 G at 100 kHz, and receiver gain = 1 x 10^{-3}.

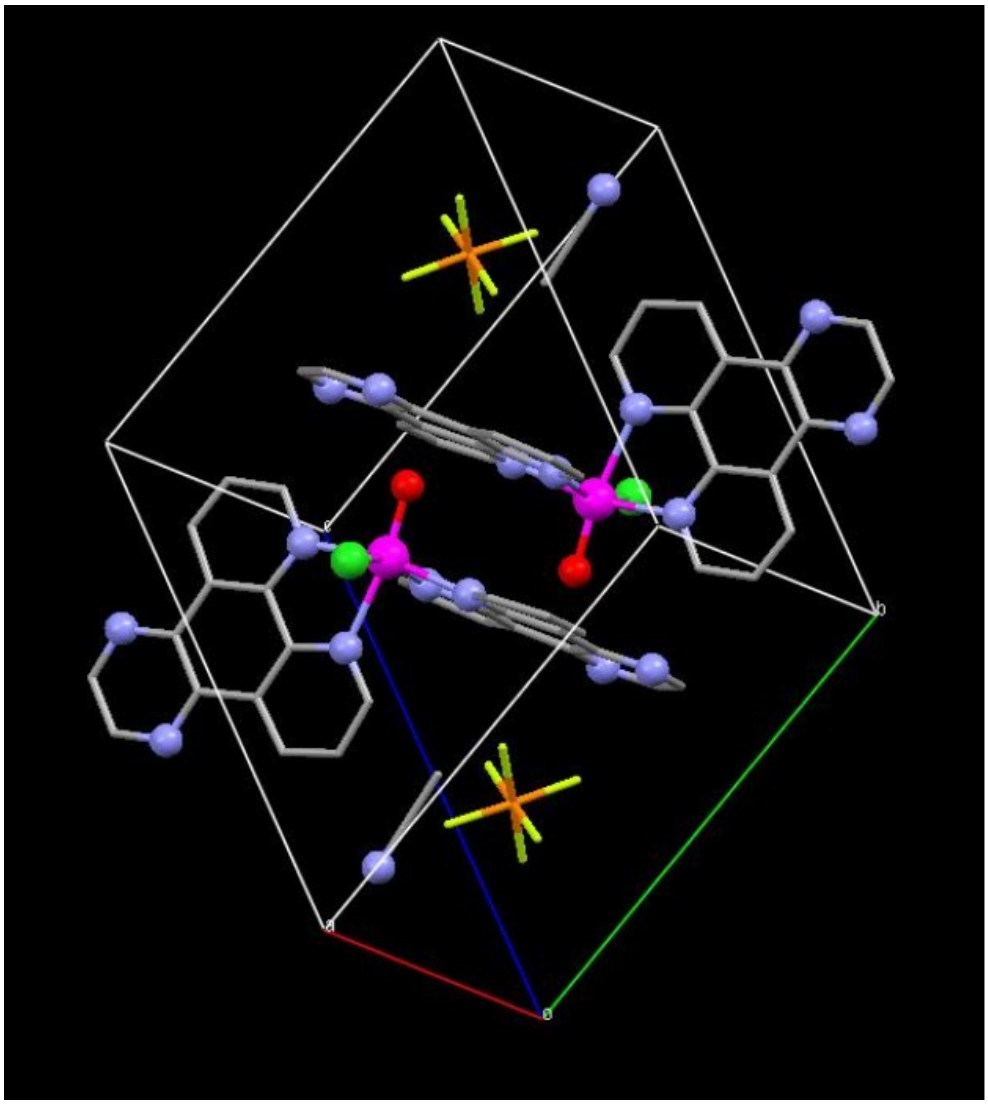

Fig. S6. Unit cell packing diagram of $[VOCl(dpq)_2](PF_6)\cdot MeCN$ (**2a**·MeCN).

Fig. S7. The energy-minimized structure of the dppz complex **3** generated using the atomic coordinates of $[VOCl(dpq)_2](PF_6)$ (**2a**·MeCN).

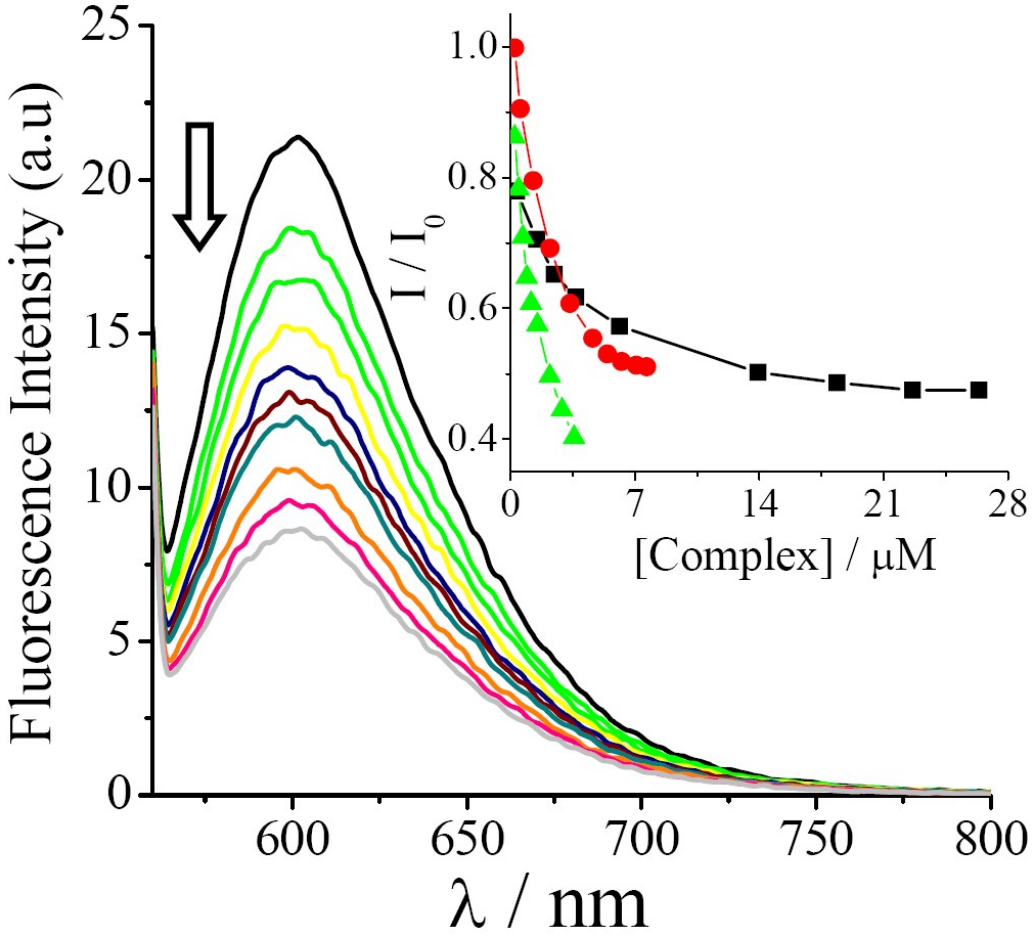

Fig. S8. Emission spectral traces of complex **3** using 350 μM calf thymus DNA-bound ethidium bromide (1.3 μM) at different complex concentrations in 5 mM phosphate buffer (pH, 6.85) at 25 ºC. The inset shows the plots for the complexes **1** (■), **2** (●) and **3** (▲).

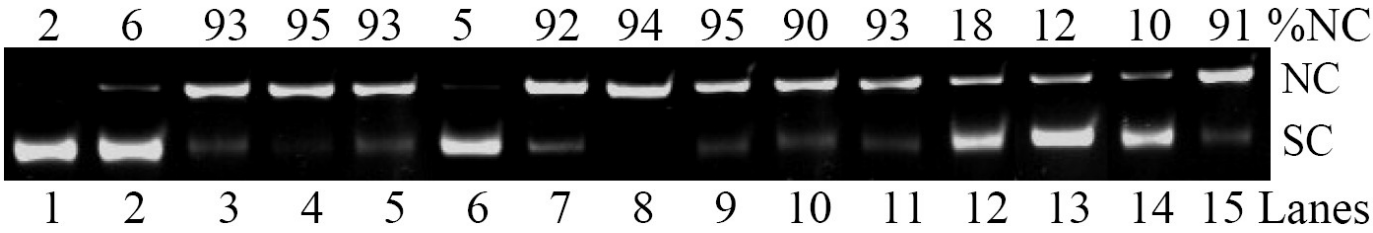

Fig. S9. Cleavage of SC pUC19 DNA (0.2 μg, 30 μM) by $[VOCl(B)_2]Cl$ (B = phen, **1**; dpq, **2**; dppz, **3**) in the presence of MPA (0.2 mM) and GSH (0.2 mM) as reducing agents in 50 mM Tris-HCl/NaCl buffer (pH 7.2) containing 1.5% DMF under dark reaction condition. Detail conditions are given below in a tabular form. The inhibition of cleavage was observed in the presence of hydroxyl radical scavengers like DMSO, KI and catalase.

Lane No	Reaction conditions
1	DNA control
2	DNA + MPA
3-5	DNA + complexes **1-3** + MPA
6	DNA + GSH
7-9	DNA + complexes **1-3** + GSH
10	DNA + complex **3** + MPA + NaN_3 (0.2 mM)
11	DNA + complex **3** + MPA + TEMP (0.2 mM)
12	DNA + complex **3** + MPA + DMSO (4 μL)
13	DNA + complex **3** + MPA + KI (0.2 mM)
14	DNA + complex **3** + MPA + catalase (4 units)
15	DNA + complex **3** + MPA + SOD (4 units)

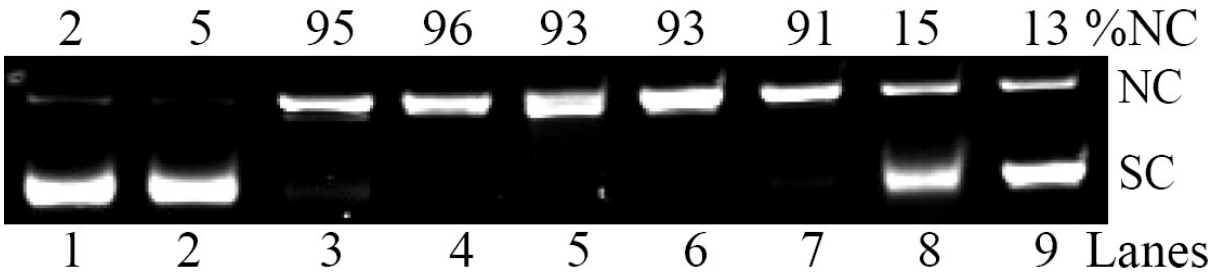

Fig. S10. Cleavage of SC pUC19 DNA (0.2 μg, 30 μM) by $[VOCl(B)_2]Cl$ (B = phen, **1**; dpq, **2**; dppz, **3**) (10 μM) in the presence of H_2O_2 (0.2 mM) as an oxidizing agent in 50 mM Tris-HCl/NaCl buffer (pH 7.2) containing 1.5% DMF under dark reaction condition. Detail conditions are given below in a tabular form.

Lane No	Reaction conditions
1	DNA control
2	DNA + H_2O_2
3-5	DNA + complexes **1-3** + H_2O_2
6	DNA + **3** + H_2O_2 + NaN_3 (0.2 mM)
7	DNA + **3** + H_2O_2 + TEMP (0.2 mM)
8	DNA + **3** + H_2O_2 + DMSO (4 μL)
9	DNA + **3** + H_2O_2 + KI (0.2 mM)

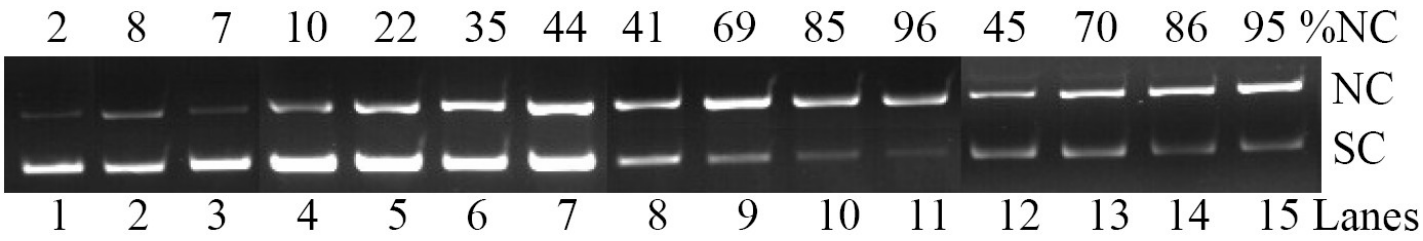

Fig. S11. Cleavage of SC pUC19 DNA (0.2 μg, 30 μM) by the complexes **1** – **3** (2.5 μM) in 50 mM Tris-HCl/NaCl buffer (pH, 7.2) containing 1.5% DMF on photo-irradiation at 365 nm (6 W) using different exposure times. Detail conditions are given below in a tabular form.

Lane No	Reaction conditions
1	DNA control (60 min)
2	DNA + dpq (10 μM) (60 min)
3	DNA + dppz (10 μM) (60 min)
4-7	DNA + complex **1** (15, 30, 45, 60 min.)
8-11	DNA + complex **2** (15, 30, 45, 60 min.)
12-15	DNA + complex **3** (15, 30, 45, 60 min.)

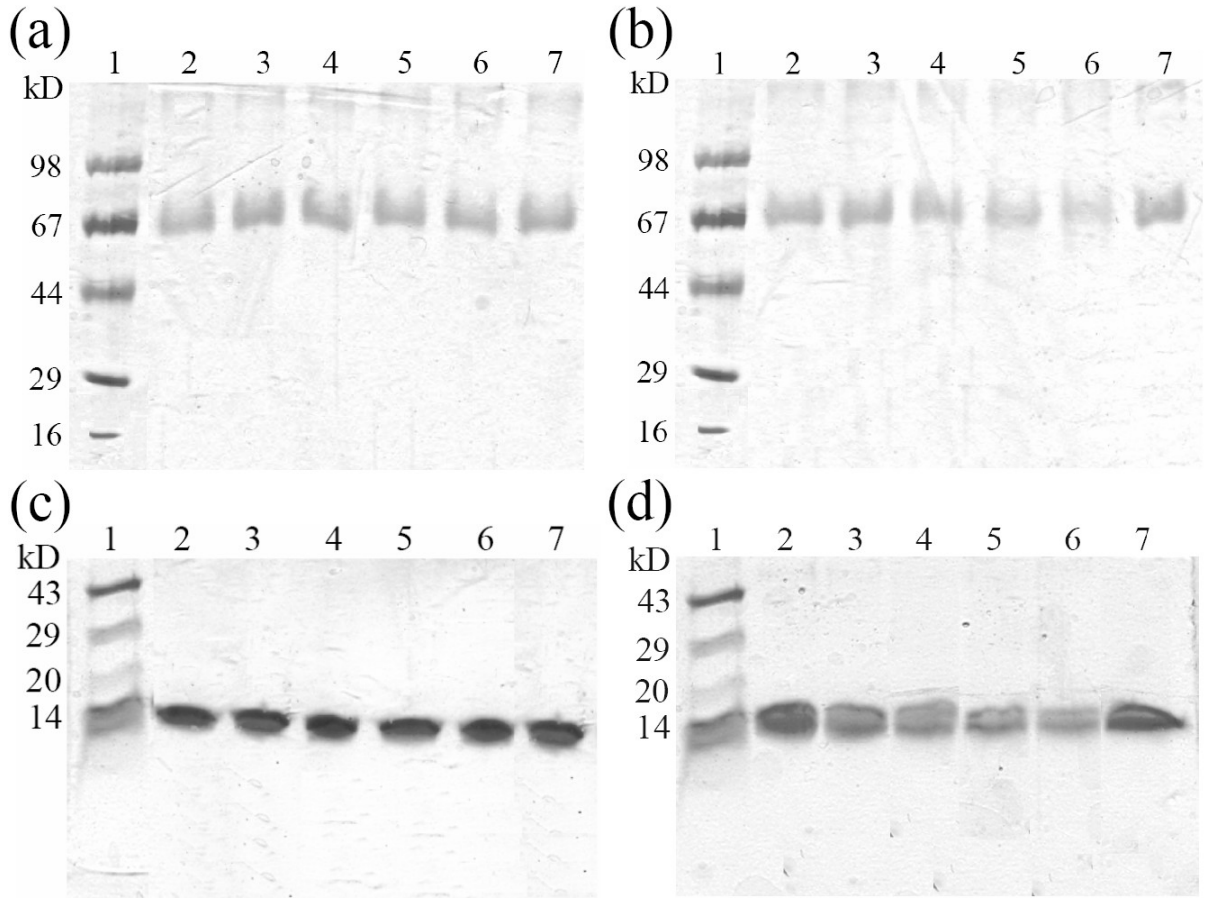

Fig. S12. Photo-induced cleavage of bovine serum albumin (BSA, 5 μM) and lysozyme (10 μM) in UV-A light of 365 nm by the complexes $[VOCl(B)_2]Cl$ (B: phen, **1**; dpq, **2**) in 50 mM Tris-HCl buffer having 0.75% DMF (pH 7.2). Panels (a) and (b) represent complexes **1** and **2**, respectively, in 12.5% SDS-PAGE: lane 1, molecular marker; lane 2, BSA control; lane 3, BSA + complex (25 μM, 2 h); lane 4, BSA + complex (50 μM, 2 h); lane 5, BSA + complex (75 μM, 2 h); lane 6, BSA + complex (100 μM, 2 h); lane 7, BSA + complex (100 μM, in dark). Panels (c) and (d) represent complexes **1** and **2**, respectively, in 16.5% tricine-SDS-PAGE: lane 1, molecular marker; lane 2, lysozyme control; lane 3, lysozyme + complex (25 μM, 2 h); lane 4, lysozyme + complex (50 μM, 2 h); lane 5, lysozyme + complex (75 μM, 2 h); lane 6, lysozyme + complex (100 μM, 2 h); lane 7, lysozyme + complex (100 μM, in dark).

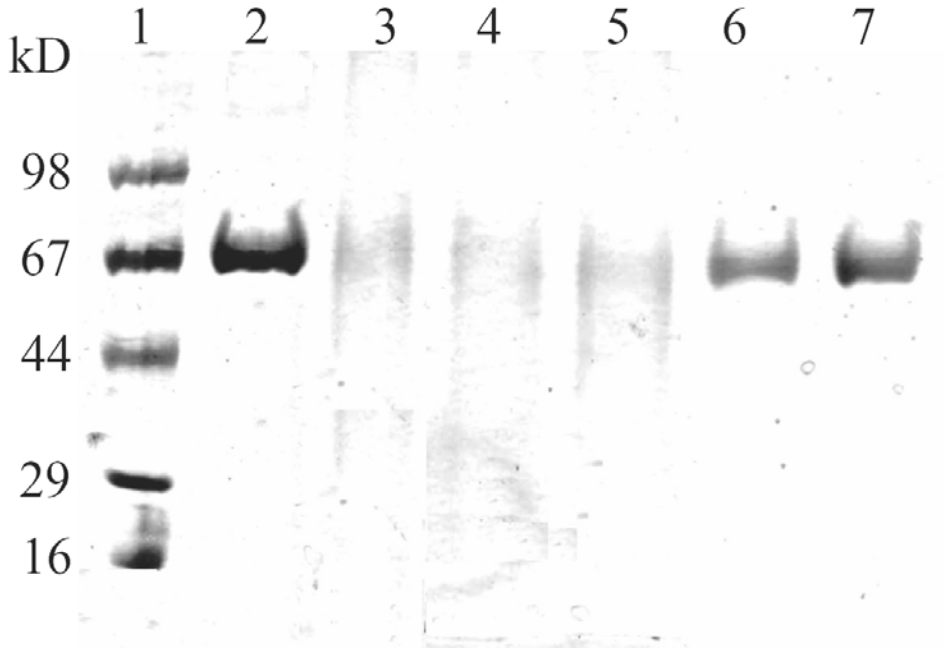

Fig. S13. 12.5% SDS-PAGE of BSA (5 μM) on 2 h photoexposure in UV-A light (365 nm, 6 W) using complex **3** (100 μM) in the presence of different additives in a 0.75% DMF-Tris-HCl buffer (50 mM, pH 7.2) at 25 °C: lane 1, standard molecular weight marker; lane 2, protein control on photoexposure; lane 3, BSA + **3**; lane 4, BSA + **3** + NaN_3; lane 5, BSA + **3** + TEMP; lane 6, BSA + **3** + DMSO; lane 7, BSA + **3** + KI.

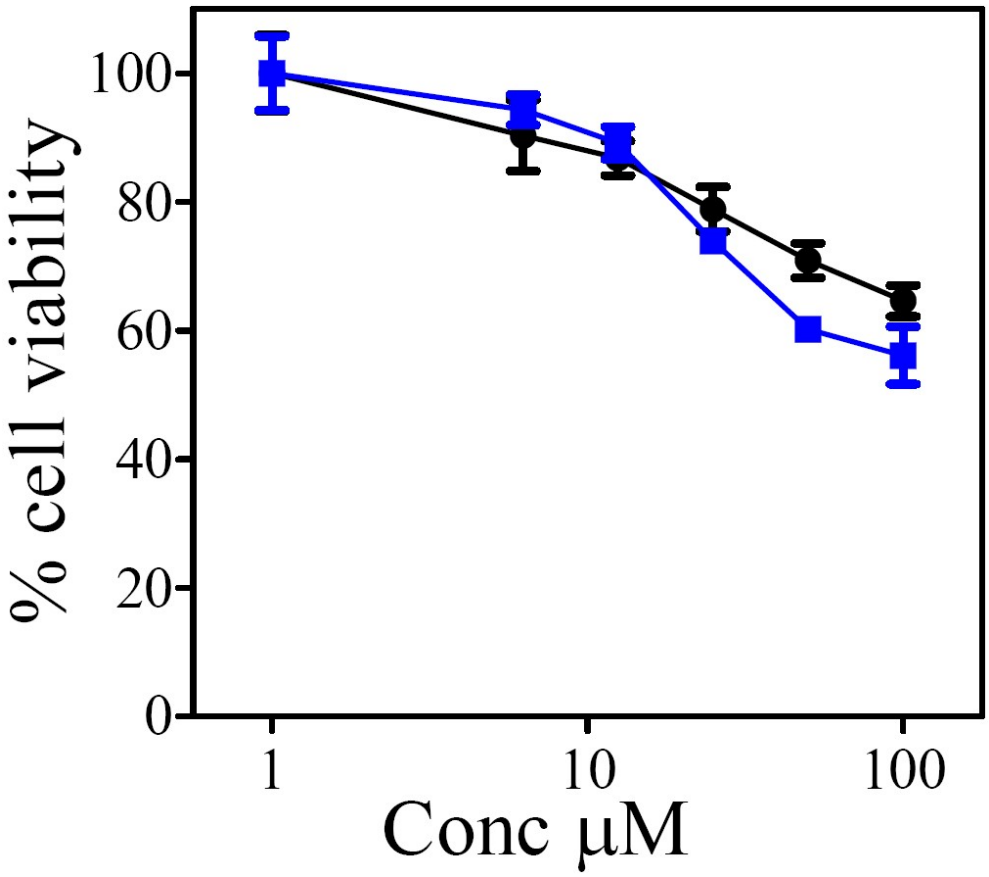

Fig. S14. Photocytotoxicity of the $[VOCl(dpq)_2]Cl$ (**2**) in HeLa cells upon incubation for 30 min in dark followed by irradiation with UV-A light (365 nm, 0.549 J cm^{-2}) as determined by MTT assay. The dark-treated cells are shown by circle (●) and the UV-A light-exposed cells are shown by squares (■). The IC_{50} values in dark and UV-A light are greater than 100 µM.

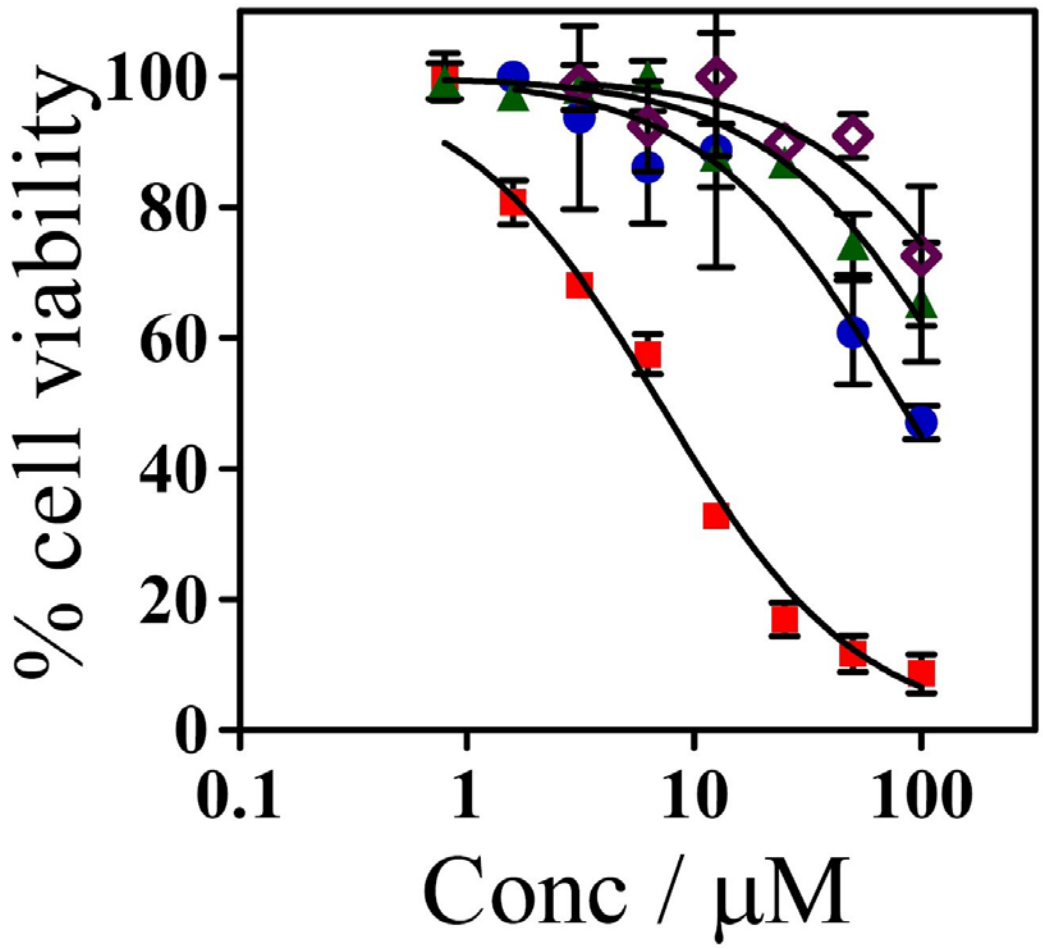

Fig. S15. Photocytotoxicity of $[VOCl(dppz)_2]Cl$ (**3**) (■), dppz ligand (●), dpq ligand (▲) and $VOSO_4$ (◊) in HeLa cells upon incubation for 30 min in dark followed by irradiation with visible light (400 to 700 nm, 20 J cm^{-2}) as determined by MTT assay. The IC_{50} values are 12, 95, >100 and >100 μM, respectively.

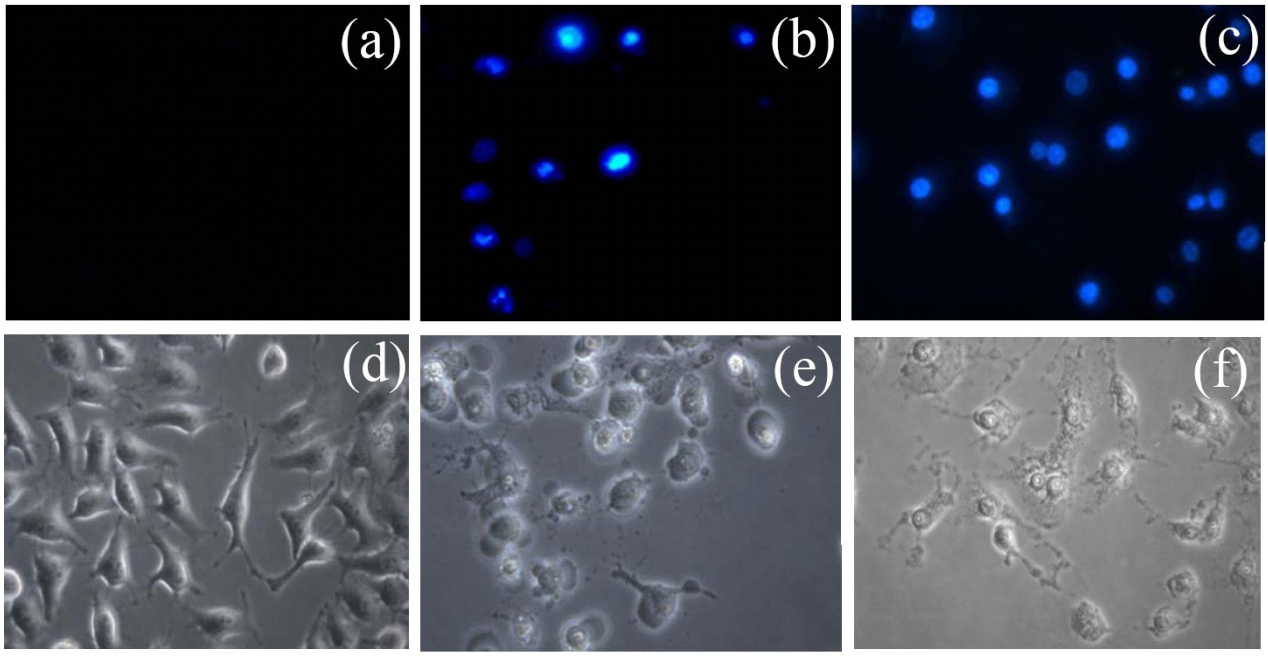

Fig. S16. DAPI staining of nuclei of HeLa cells (2 h post photoexposure) treated with **3** and UV-A light of 365 nm: (a) untreated control cells, (b) cells treated with 1 μM of **3** and (c) cells treated with 10 μM of **3**. Panels (a)-(c) are from a fluorescence microscope with 360/40 nm excitation filter and 460/50 nm emission filter. Panels (d)-(f) are the respective bright-field images.