Electronic Supplementary Material (ESI) for ChemComm.

Supporting Information

A Versatile Water-Soluble Chelating and Radical Scavenging Platform **

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Experimental part

Dry distilled CH_2Cl_2, CH_3CN and CH_3OH were purchased from Aldrich Chemical Ltd. Commercially available, solid organic chemicals were purchased from Fluka, Acros and Aldrich and used without further purification. Copper acetate monohydrate was purchased from Acros organic. Ammoniumiron(II)sulfatehexahydrate (99%) and 5,5-dimethyl-1- pyrroline *N*-oxide (DMPO) was purchased from Aldrich–Sigma (Israel). Hydrogen peroxide (30 %) was purchased from BioLab (Israel). Dulbecco's modified Earle's medium (DMEM), heat inactivated fetal calf serum (FCS), L-glutamine, penicillin/streptomycin, neomycine-G418, trypsin, were obtained from Biological Industries (Beit Haemek, Israel). Glucose oxidase, Tris-Cl, Trichloroacetic acid (TCA), PBS tablets, DMSO, Bradford reagent, peroxidase, ferrous ammonium sulfate, sodium thiocyanate reagent were purchased from Sigma-Aldrich (St. Louis, MO, USA), Thiazolyl Blue Tetrazolium Blue (MTT) was from Chem-Impex International (Wood Dale, IL, USA), Hygromycine B and 2,2-Azino-di-[3-ethylbenzthiazoline sulfonate] (ABTS) were purchased from Molecula (Shaftesbury, UK), Metmyoglobin and total antioxidant measurement kit were from Cayman Chemical Company, (Ann Arbor, USA).

Ligand synthesis.

bis-(2,5-dioxopyrrolidin-1-yl) 5-(-butyl)-2-methoxyisophthalate. 5-(*tert*-butyl)-2-methoxyisophthalic acid (5 g, 19 mmol) and N-Hydroxysuccinimide (5.02 g, 43 mmol) were dissolved in 80 mL of dioxane under N_2 atmosphere. A solution of 1,3-dicyclohexylcarbodiimide (9 g, 43 mmol) in 85 mL of CH_2Cl_2 was added dropwise to the reaction mixture at 10 ^oC. The resulting suspension was stirred at room temperature for 12 h, filtered and the filtrate was evaporated. The solid was recrystallised from hot ethanol to give a white powder in 47% yield (4.17 g). ^1H NMR (300 MHz, $CDCl_3$, 25 ^oC): δ/ppm 1.36 (s, 9H, ^{t}Bu), 2.91(s, 8H, CH_2), 4.02 (s, 3H, CH_3), 8.2 (s,2H, ArH). ^{13}C NMR (75 MHz, $CDCl_3$, 25 ^oC): δ/ppm 25.7, 31 (CH_3, ^{t}Bu), 34.77 (C-, ^{t}Bu), 64.43 (CH_3-O), 121.22 (Ar-C), 134.79 (Ar-Cmeta), 147.43 (Ar-C), 159.81(C-OMe, Ar), 160.4 (C(O)O), 169.98 (C(O)N).

*5-(tert-butyl)-N1,N3-bis(2-hydroxyethyl)-2-methoxyisophthalamide (**1OMe**).* 2-aminoethanol (1.64 g, 27 mmol, 1.62 cm^3) was dissolved in 10 cm^3 of CH_2Cl_2 under N_2 atmosphere. A solution of **P** (3 g, 6.7 mmol) in 80 cm^3 of CH_2Cl_2 was added dropwise to the reaction mixture at room temperature. The resulting suspension was stirred at room temperature for 12 h, filtered and the filtrate was evaporated and recrystallised from acetone/hexane to give **1OMe** as a white powder in 77% yield (1.76 g). ^{1}H NMR (300 MHz, $CDCl_3$, 25 ^{o}C): δ 1.205 (s, 9H, ^{t}Bu), 3.46 (t, 4H, CH_2, J = 5.1 Hz), 3.66 (t, 4H, CH_2, J = 5.1 Hz), 3.72 (s, 3H, OCH_3), 7.92 (s,2H, Ar H), 8.032 (t, 2H, NH). EI-MS: m/z 339 ($[M-H]^+$, 100%).

*5-(tert-butyl)-2-hydroxy-N^1,N^3-bis(2-hydroxyethyl)isophthalamide (**1OH**).* **1OMe** (1.84 g, 5.4 mmol) was dissolved in 150 cm^3 of distilled CH_2Cl_2 under N_2 atmosphere. BCl_3 1 M in CH_2Cl_2 (2.23 g, 19 mmol**,** 19 cm^3) was added dropwise to the reaction mixture at 10 ^{o}C and stirred for 72 h. Then, methanol (100 cm^3) was slowly added and the solution was stirred for an hour. The solvent was evaporated, recrystallised from hot water and filtered to give **1OH** as a white powder in 73% yield (1.29 g). ^{1}H NMR (300 MHz, D_2O, 25 ^{o}C): δ 1.32 (s, 9H, ^{t}Bu), 3.58 (t, 4H, CH_2, J=5.5Hz), 3.79 (t, 4H, CH_2, J=5.5 Hz), 8.03 (s,2H, Ar H). Anal calc for $C_{16}H_{24}N_2O_5$: C 59.24, H 7.46, N 8.64 Found: C 58.81, H 7.73, N 8.07. EI-MS: m/z 325 ($[M-H]^+$, 100%).

bis(2-aminoethyl) 5-(tert-butyl)-2-hydroxyisophthalate tetrabutyl ammonium salt*, **([1O][NBu$_4$])**.* To a solution of **1OH** (0.432 mmol, 140 mg) in dry methanol (5 cm^3), was added a 1 M solution in methanol of tetrabutylammonium hydroxide (0.432 cm^3, 0.432 mmol). The solution was stirred under N_2 atmosphere for 2 h. Then the solvent was removed under vacuum yielding an orange oily material which was recrystallized from methanol/ diethyl ether yielding colourless crystalline powder of **[1O][NBu$_4$]** in 92 % yield (0.22 g)**.** Single crystals suitable for X-ray crystallography were obtained in this manner. MS-EI(-): m/z 889 ($\{[1OMe]_2[NBu_4]\}^-$ 100%). ^{1}H NMR (300 MHz, CD_3CN, 25 ^{o}C): δ 0.96 (t, 12H, CH_3-, [NBu_4], J = 7.3Hz), 1.26 (s, 9H, ^{t}Bu), 1.34 (m, 8H, -CH_2-, [NBu_4]), 1.58 (m, 8H, -CH_2-, [NBu_4]), 3.06 (m, 8H, -CH_2-, [NBu_4]), 3.4 (m, 4H, CH_2), 3.6 (bs, 4H, CH_2), 8.04 (bs,2H, ArH), 12.26 (bs, 2H, NH). ^{13}C NMR (300 MHz, CD_3CN, 25 ^{o}C): 13.78 (-CH_3, [NBu_4]), 20.32 (-CH_2-, [NBu_4]) 24.28 (-CH_2-, [NBu_4]) 32.06 (CH_3, ^{t}Bu), 43.15 (CH_2-NH), 59.3 ($^+$N-CH_2-,[NBu_4], 1:1:1, t, J = 3 Hz), 64 (CH_2-OH), 120.9

(Ar-C), 131.29 (Ar-C_{meta}), 131.74 (Ar-C), 171.57(C(O)NH). Anal calc for $C_{32}H_{59}N_3O_5$: C 67.93, H 10.51, N 7.43 Found: C 67.54, H 10.38, N 7.83. IR **(**ZnSe**):** υ b3290(NH), 1645 (C=O) cm^{-1}.

Complex synthesis.

[Cu(1O)$_2$(H$_2$O)]. To a solution of **1OH** (0.1 g, 0.3 mmol) in methanol (6 mL), a solution of Cu(acetate)$_2$·H_2O (0.03 g, 0.15 mmol) in methanol (8 mL) was added. Then, Et_3N (1.3 mL) was added to the reaction mixture and the reaction was left overnight which afterword was evaporated to give green oil. The oil was washed several times with diethyl ether, evaporated to dryness and crystallized from methanol/diethyl ether in liquid/liquid diffusion to give green rectangular crystals of **[Cu(1O)$_2$(H$_2$O)]** suitable for X-ray crystallography were collected in 74% yield (0.079 g, 0.11 mmol). MS ES(+): m/z 710 $(M+H)^+$. Elemental analysis: Calc. For $C_{32}H_{46}N_4O_{10}Cu·H_2O$: C 52.9, H 6.34, N 7.72; Found: C 52.64, H 6.34, N 7.40. UV/vis (EtOH): λmax/nm (ε/M^{-1} cm^{-1}): 290 (sh) (7233), 348 (12395), 425(sh) (764), 696 (77).

[Fe(1O)$_3$]. To a solution of **1OH** (0.1 g, 0.3 mmol) in methanol (6 mL), a solution of Fe(ClO_4)$_3$·H_2O (0.038 g, 0.1 mmol) in methanol (8 mL) was added. Then, Et_3N (1.26 mL) was added to the reaction mixture and the reaction was left overnight which afterword was evaporated to give red oil. The oil was washed several times with diethyl ether, evaporated to dryness and crystallized from methanol/diethyl ether in liquid/liquid diffusion to give red crystals of **[Fe(1O)$_3$]** 70% yield (0.071 g, 0.07 mmol). MS ES(+): m/z 1026 $(M+H)^+$. Elemental analysis: Calc. For $C_{48}H_{69}FeN_6O_{15}$: C 56.19, H 6.43, N 8.19; Found: C 55.81, H 6.75, N 7.73. UV/vis (EtOH): λmax/nm (ε/M-1 cm-1): 290 (sh) (12962), 335 (19085), 458 (4231). The X-band EPR spectrum in frozen shows a rhombic S = 5/2 octahedral spectral with a g values at 4.3.

Physical methods. Elemental analyses of the compounds isolated in these studies were accomplished using the chemistry departmental service (BIU). EI mass spectra were recorded on a Q-Tof micro (UK)-micromass-waters spectrometer. ^1H and ^{13}C NMR spectra were recorded on Bruker DPX300 NMR and Bruker Avance III 400 spectrometer

operating at 300 and 400 MHz, respectively for ^{1}H and 75 and 100.62 MHz, respectively for ^{13}C. Chemical shifts are reported in ppm downfield from tetramethylsilane, and coupling constants (*J*) are reported in Hz. Resonance and structural assignments were based on the analysis of coupling patterns, including the ^{13}C-^{1}H coupling profiles obtained in bidimensional heteronuclear multiple bond correlation (HMBC) and heteronuclear single quantum coherence (HSQC) experiments, performed with standard pulse programs. X-band EPR spectra were obtained at 120-130K K on a Bruker ELEXSYS E500 spectrometer equipped with a Bruker ER4131VT variable-temperature unit. Samples were frozen in 3 mm quartz tubes and placed into a standard rectangular Bruker EPR cavity (ER 4119 HS)., The spectra were recorded at a microwave frequency of ~9.3 GHz , a microwave power of 6.3 mW and modulation amplitude of 5 G. All EPR spectra shown have been background-subtracted from empty tube blank with subsequent base-line correction using the XEPR software package. The spectra were simulated using Matlab with easy spin (5.0.2 (http://www.easyspin.org/)) toolbox.

UV/Vis spectra were recorded on a Varian Cary 5000 UV/Vis /NIR spectrophotometer. The measurements were carried out using a quartz cuvette with optical path length 0.1 cm. IR spectra were recorded on a Nicolet iS10 FT-IR spectrometer using ATR accessory on ZnSe crystal for powder samples pressed. Add CV details.

X-ray crystallography. Single crystal X-ray diffraction data for compounds **1OH** and **[Cu(1O)$_2$(H$_2$O)]** were collected on a Bruker SMART CCD 6000 area detector diffractometer and for **[1O][NBu$_4$]** on a Rigaku R-AXIS SPIDER IP diffractometer (ω-scan, graphite-monochromated λMoKα radiation, λ=0.71073Å). All data were collected at 120.0(1)K maintained bythe Cryostream (Oxford Cryosystems) open-flow nitrogen cryostats. All structures were solved by direct methods and refined by full-matrix least squares on F^2 for all data using OLEX2 [1] and SHELXTL[2] software. All non-disordered non-H atoms were refined with anisotropic atomic displacement parameters, disordered atoms were refined with various fixed SOF. The hydrogen atoms in the structure **[Cu(1O)$_2$(H$_2$O)]** and of terminal and disordered groups in other structures were placed in calculated positions and refined in "riding" mode.

The unit cell, data collection and refinement parameters for **1OH, [1O][NBu_4]** and **[$Cu(1O)_2(H_2O)$]** are given in **Table S1**. The CCDC entries 829301, 829302 and 995321 contain the supplementary crystallographic data for this paper. These data can be obtained free of charge from the Cambridge Crystallographic Data Centre *via* www.ccdc.cam.ac.uk/data_request/cif.

UV photolysis of H_2O_2. To estimate the scavenging abilities of our compounds against •OH, we have used aa spin trap electron spin resonance (EPR) method where the hydroxyl radical was generated from UV photolysis of H_2O_2 as was used also by Bolton JR etal [3] and Ozawa T [4] .

photolysis

H_2O_2 --------> 2 •OH

DMPO + •OH -------> DMPO-OH

The reaction mixture containing DMPO (10 mM) and H_2O_2 (2.5 mM) in purified water (total volume 100 µl) and different concentration of antioxidant. The final volume of the reaction mixture, 100 µl was drawn by a syringe into a gas-permeable Teflon capillary (Zeus, Raritan, NJ). Than the capillary was folded twice and inserted into a narrow quartz tube that was open at both ends. This quartz tube was placed in a 3VL-315 365 nm UV lamp, Vilber Lourmat, France and irradiation with energy of 7.2 J/cm^2. After irradiation the quartz tube was placed immediately at the EPR cavity and measured. The measurements were performed with a Bruker ER 100d X-band spectrophotometer, using microwave frequency, 9.76 GHz; modulation frequency, 100 kHz; microwave power, 20 mW; modulation amplitude, 1 G; time constant, 655.36 ms; sweep time, 83.89 s; and receiver gain, 1×10^5 .

The DMPO-OH quartet signal intensities were evaluated by the peak to peak height of the second low field peak of the DMPO–OH spin adduct quartet. The scavenging values are expressed as percentage of antioxidant inhibition , where the concentration of antioxidants that causes a 50 % decrease in the intensity of DMPO-OH (50 % inhibitory concentration where defined as IC50.

% antioxidant inhibition α % of DMPO-OH quenching= $\frac{I_{control} - I_{antioxidant}}{I_{control}} \times 100$

$I_{control}$ is the DMPO-OH intensity in the absence of antioxidant and $I_{antioxidant}$ is the DMPO-OH intensity with addition of antioxidant

Cell culture. Stably transfected NSC-34 cells by human G93A SOD1 gene (^{G93A}hSOD1-NSC-34) were kindly donated by Prof. Nava Zisapel (Tel Aviv University, Israel). Cells were grown in DMEM (22.5 mM glucose) supplemented with 20% heat inactivated fetal calf serum (FCS), 1 mM glutamine, and antibiotics (100 μg/mL penicillin, 100 μg/mL streptomycin, 200 μg/mL hygromicine and 700 μg/mL neomycin-G418) at 37° C in a 5% CO_2 humidified atmosphere [5]. L6 myotubes were obtained through the courtesy of Prof. Shlomo Sasson (Hebrew Jerusalem University, Israel), and were cultured as previously described [6].

Metmyoglobin/ABTS/H_2O_2 assay. Solutions with increasing concentrations of Trolox, **1OH**, **2OH** and **1OMe** (50 μM - 330 μM) in potassium phosphate buffer (5 mM potassium phosphate, pH 7.4, 155 mM sodium chloride and 5.5 mM glucose) were placed in a 96 wells plate. Metmyoglobin (0.047 μM, final concentration) and ABTS (0.22 mM, final concentration) dissolved in potassium phosphate buffer were added to all wells; after which H_2O_2 (20 μM final concentration) was rapidly supplied to each well. After 30 min incubation at room temperature on slow agitation, absorption of the $ABTS^{\bullet+}$ was measured on a plate reader at λ = 750 nm.

Biological assays. In all experiments, the tested compounds were initially dissolved in DMSO to prepare the stock solutions. The maximal concentration of DMSO in cell medium was 12.5 mM. Same amount of DMSO was present in the control solution, to insure valid comparison. In addition, independent experiments have shown that DMSO had no significant effect on these assays at this concentration.

Cytotoxicity test. Rat phaeochromocytoma cell line, PC-12, was maintained routinely in low glucose DMEM supplemented with 10% horse serum and 5% FBS, 2 mM L-

glutamine, 100 U/ml penicillin and 100 mg/ml streptomycin in 5% CO2 atmosphere at 37°C.

In order to determine the toxicity of the compounds, PC-12 cells or L6 myoblasts (10,000 cells/well) were plated in 96-well tissue culture plates in 0.1 mL of the medium and incubated for overnight for attachment. The medium was then replaced with fresh medium containing a serial dilution of tested compounds starting from 100 μM. Following 24 h incubation of the plates at 37°C, the cell survival was determined by MTT assay. The compounds have shown no sign of toxicity even at 100 μM.

Glucose Oxidase/glucose oxidative stress generating system: determination of H_2O_2 concentration. Concentration of H_2O_2 generated by glucose oxidase/glucose system was determined as described by Thurman et al [7]. The ^{G93A}hSOD1-NSC-34 or ^{wt}hSOD1-NSC-34 cells were incubated in growing medium (without phenol red) containing glucose (23.5 mM) with 50 mU/ml glucose oxidase. After three different periods of time (1, 2 and 4 hours) aliquots 1 mL of medium were collected, and 0.05 ml TCA (0.27 mM, final concentration) was added. The samples were centrifuged at 500 g for 10 min, 0.2 ml of 10 mM ferrous ammonium sulfate (1.67 mM, final concentration) and 0.1 ml of 2.5 M sodium thiocyanate were added to the supernatant (0.17 M, final concentration). Absorption of the ferrithiocyanate complex formed was measured at λ = 480 nm, and compared to standard obtained from dilutions of a standard H_2O_2 concentrations.

MTT cell viability assay. This assay measures the reduction of a tetrazolium component in MTT (3-(4,5-dimethythiazol-2-yl)-2,5-diphenyl tetrasodium bromide] into an insoluble formazan produced by the mitochondria of viable cells. Cells were grown as described above. Glucose oxidase (50 mU/ml) was added to cell medium containing glucose (23.5 mM) to generate oxidative stress. After 4 hours incubation time, the tested compounds (100 μM) were added to cells medium for 1 hour incubation time, after which time MTT (4.8 mM, final concentration) was supplied. Cells were further incubated at 37 °C for 2 hours. The medium was then aspirated and the cells were washed using PBS, and then solubilized in 200 μl of DMSO. Absorbance at 570 nm and subtract background at 670 nm, in 96 wells plate (100 ul per well) was measured in an ELISA reader. The intensity

of the absorption band at 570 nm is directly proportional to the amount of viable cells. The obtained results were normalized according to total protein amount in each treatment. Total protein amount was determined by Bradford method [8].

Lipid Peroxidation MDA (malondialdehyde) assay.

Lipid peroxidation was measured using the Lipid Peroxidation MDA (malondialdehyde) Assay Kit (Abcam, Cambridge, UK). The assay measures the amount malondialdehyde (MDA) produced as the end product of lipid peroxidation from lipids in cell membrane under oxidative stress conditions. The MDA is indirectly detected colorimetric measurements, by reacting it with thiobarbituric acid (TBA) generating the MDA-TBA adduct which is detected by at 532 nm.

^{G93Ah}SOD1-NSC-34 cells were grown as described above. Trolox , **1OH** and **1OMe** (100 μM) were added in DMSO (0.5% concentration) to the cultures. After 30 min oxidative stress was induced by GO system as described above. GO-untreated cells were used as a control. After 1 h, the cells (2 x 10^6 cells) were gently washed with cold PBS three times and detached by trypsin, and lysed using a lysis solution containing 300 μL of the MDA lysis buffer with 3 μL BHT (5% Butylated hydroxytoluene). Homogenization of cells was achieved by using a Dounce homogenizer (10 passes) on ice, until efficient lysis was obtained (confirmed by cell viewing under light microscope). Then, the lysates were collected, centrifuged for 10 min at 13,000 × g and the supernatant was used for the MDA level measurements. The reaction mixture was incubated at 95°C for 60 min with thiobarbituric acid (TBA, 600 μL/well) to generate the MDA-TBA adduct. After then the samples were cooled to room temperature on in ice bath for 10 mn; and 200 μL from each treatment was pipetted into a 96-well plate and absorbance was detected by plate reader at 532 nm.

A standard calibration curve was prepared according to the manufacturer's protocol. Each sample (200 μl) and solution of different concentrations of MDA standard (200 μl) was pipetted into a 96-well plate and absorbance was read on plate reader at 532 nm.

Total trolox equivalent antioxidant capacity assay (TEAC assay).

^{G93A}hSOD1-NSC-34 cells were grown as described above. Before the experiment, cells were incubated at 37^{o}C for 2 hours in serum-free growing medium. Trolox (1 mM), **1OH** (330 μM), **1OMe** (330 μM), or DMSO (used for the control) were added to the cells. After 1 hour incubation time, H_2O_2 (500 μM) was added, and the cells were further incubated for 30 min; after which time the cells were washed by cold PBS three times to remove remains of the medium. The last portion of PBS was aspirated by suction. Cells were scribed to cold lysis buffer (20 mM, Tris (pH 7.5),150 mM NaCl and 16.2 mM NP-40) and collected to centrifuge tubes. Lysates were treated by Vortex, three times in interval of 10 min, following three cycles frozen and defrozen in liquid nitrogen. After centrifugation (8000 g for 30 min at 4° C) cell lysates were taken to determination of formation of $ABTS^{\bullet+}$ using "Total Antioxidant kit" from Cayman Chemical Company. Thus, the resulting cell lysates were submitted to metmyoglobin/H_2O_2/ABTS antioxidant assay, and the absorption of $ABTS^{\bullet+}$ was monitored at 750 nm. TEAC equivalent was calculated according to equation below as the amount of trolox equivalents per 1mg of total protein, as described in kit manual.

$$\text{Antioxidant (mM)} = \left[\frac{\text{Sample average absorbance} - \text{(y-intercept)}}{\text{Slope}} \text{ X Delution} \right] \Big/ \text{mg of total protein}$$

Standard trolox calibration curve with known concentrations of dose dependent inhibition of ABTS oxidation was used for calculation (see Figure below). The total protein amount was determined by Bradford method [8].

Statistical Analysis

Statistical significance of results (*$p<0.05$) was calculated for all experiments using the Student's two tailed test. Results are given as mean ± SEM, for triplicate experiments.

Calibration curve of Trolox. Inhibition of ABTS oxidation

Average absorbance of $ABTS^{\bullet+}$ signal in presence of known trolox concentrations.

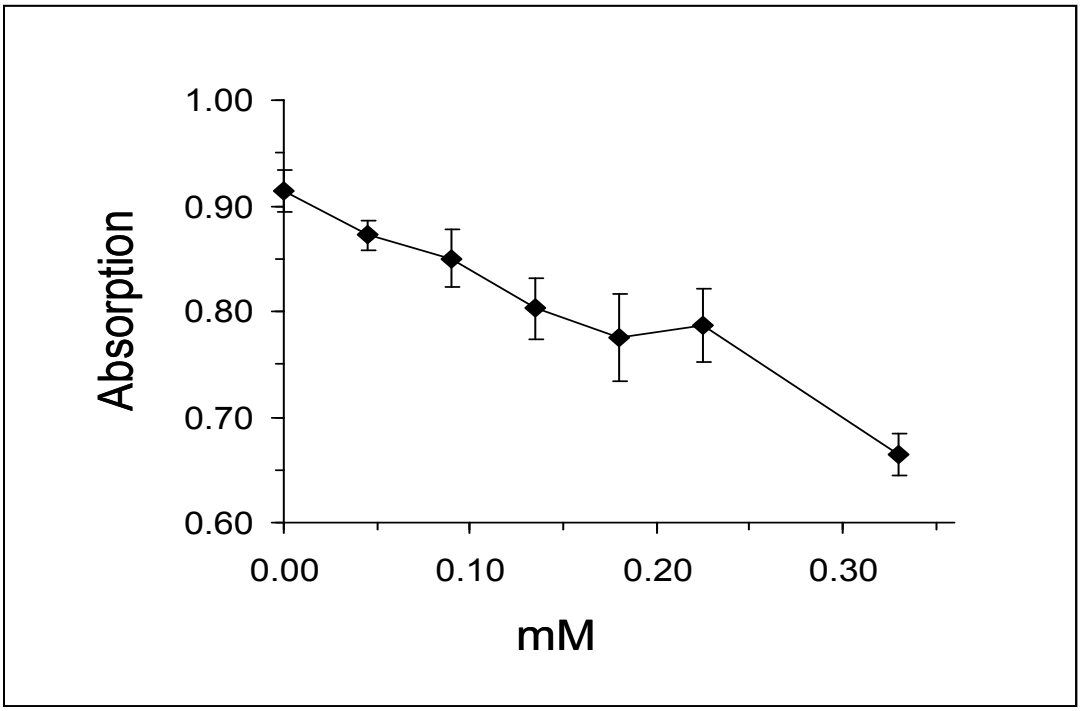

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Table S1 Crystallographic data and parameters of the refinements for **1OH**, **[1O][NBu$_4$],** and **[Cu(1O)$_2$(H$_2$O)]**.

Compound	**1OH**	**[1O][NBu$_4$]**	**[Cu(1O)$_2$(H$_2$O)]**
Empirical formula	$C_{16}H_{24}N_2O_5$	$C_{32}H_{59}O_5N_3$ x 0.5 CH_2Cl_2	$C_{32}H_{48}CuN_4O_{10}$ x H_2O
Formula weight	324.37	608.29	737.29
Crystal system	Monoclinic	Monoclinic	triclinic
Space group	$P2_1/n$	$P2_1/n$	P-1
a/Å	6.9693(2)	20.6006(6)	13.9600(6)
b/Å	12.1404(3)	12.5488(3)	14.0920(6)
c/Å	19.3065(5)	27.2830(7)	18.3587(7)
α/°	90.00	90.00	89.6370(10)
β/°	90.6230(10)	92.36(3)	83.2460(10)
γ/°	90.00	90.00	85.3260(10)
Volume/Å^3	1633.43(7)	7047.0(3)	3574.6(3)
Z	4	8	4
ρ_{calc}mg/mm^3	1.319	1.147	1.370
μ/mm^{-1}	0.098	0.149	0.675
F(000)	696	2664	1560.0
Reflections collected	17655	70244	44469
Independent reflections, R_{int}	4133, 0.0238	16157, 0.1017	18926, 0.0424
Data/restraints/parameters	4133/0/267	16157/0/794	18926/0/894
Goodness-of-fit on F^2	1.070	1.036	1.026
Final R_1 values [I>2σ (I)]	0.0661	0.0786	0.0555
Final wR_2values [all data]	0.2006	0.1778	0.1682

Figure SI1. dQ/C vs. time coulommetric plot during the electrolysis of [**1O**][NBu_4] (1 mM) in CH_2Cl_2 at 273K containing [NBu_4][PF_6] (0.1M) as supporting electrolyte. (The curve reached a plateau after 113min (dQ/C= 0.363879))

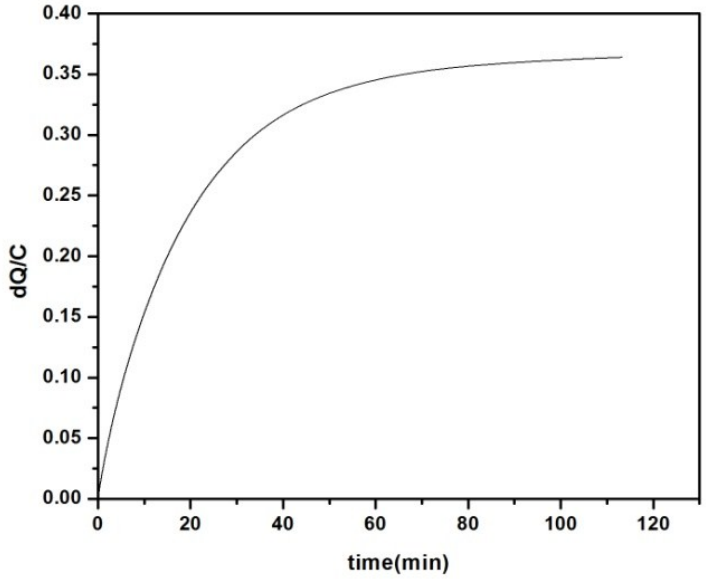
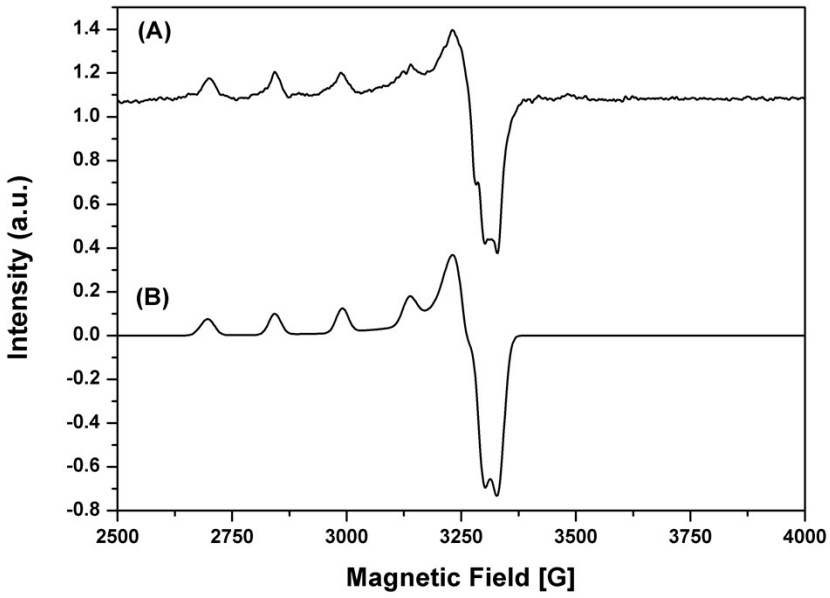


Figure SI2. X-band EPR experimental (A) and simulated (B) spectra of **1-Cu** (top) and **1-Fe** (bottom) in frozen solution (*ca.* 1 mM) in CH_3OH at 130K.

Simulation parameter for **1-Cu** : g_{zz}(2.3139) >> g_{xx}(2.0775), g_{yy} (2.0485) and $A_{zz63,65Cu}$ of 167 G ; and **1-Fe** : g = 4.2.



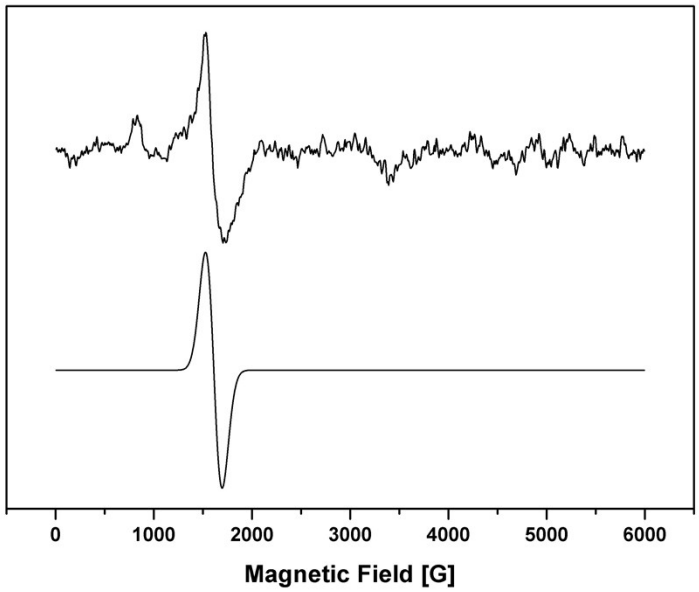


Figure SI3. ORTEP Representation of the 1D-chain $\{A/A' - B/B'\}_n$ formed in **1O-Cu**·0.5 H_2O single crystals. Intermolecular H-bonding interactions within and between the dimers A/A' and B/B' denoted by dashed lines.

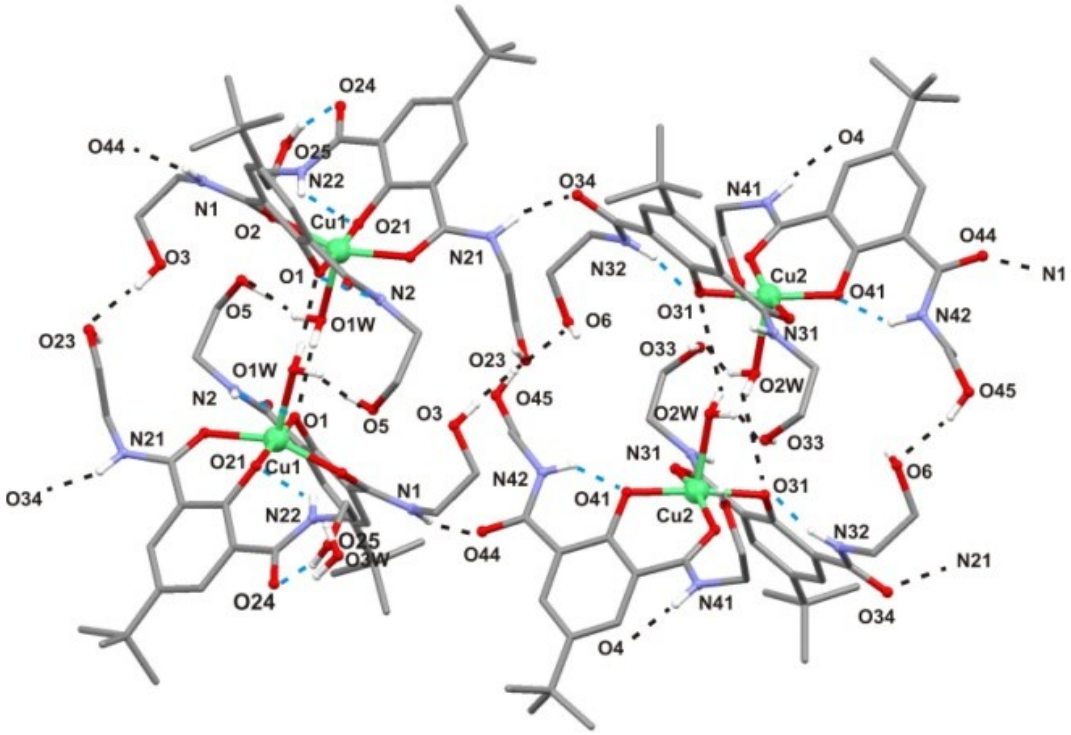
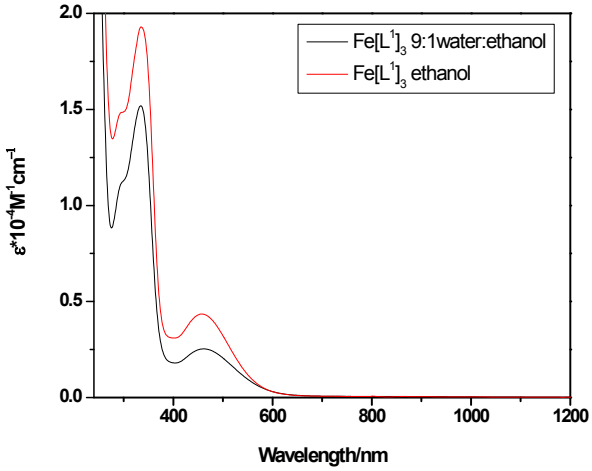


Figure SI4. UV/vis spectra of (top) **$Fe(1O)_3$** in EtOH (red line) and in 9 : 1 water/methanol (black line) solutions; and (bottom) **$Cu(1O)_2$** in EtOH (red line) and in 9 : 1 water/methanol (black line) solutions.



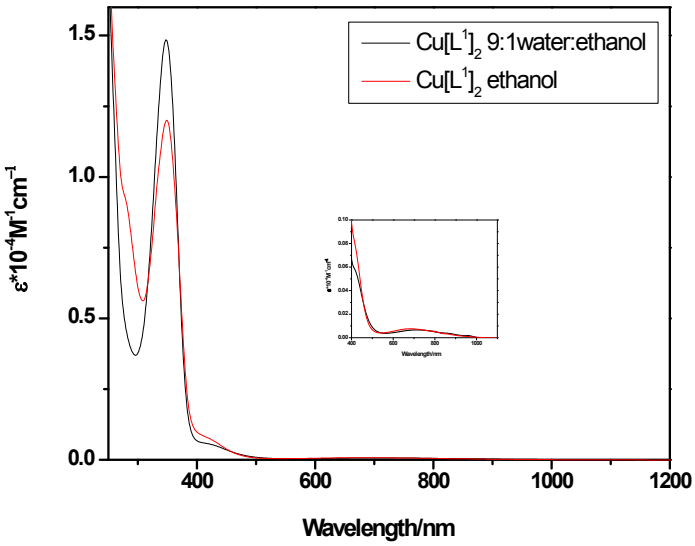


Figure SI5. UV/vis spectra of (**1OH**)/$Fe^{III}(ClO_4)_3$ mixtures in ratios from 0 to 1 in 9 : 1 water/methanol solutions.

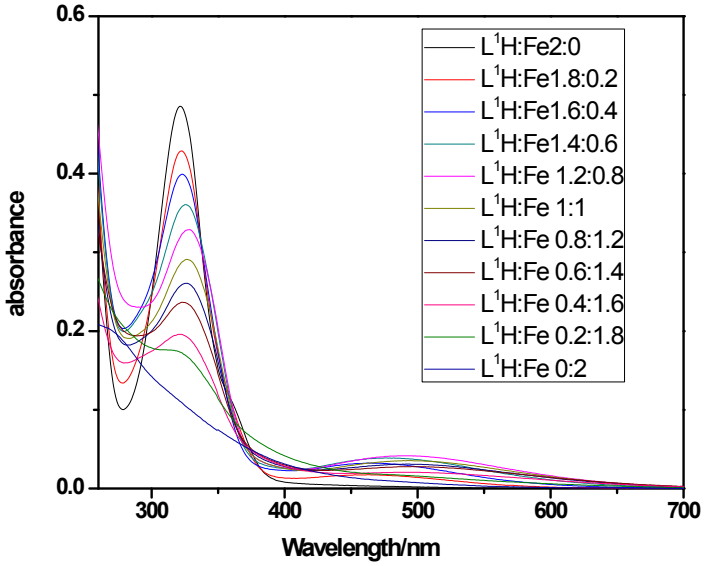


Figure SI6. UV/vis spectra of (**1OH**)/$Cu^{II}(SO_4)_2$ mixtures in ratios from 0 to 1 in 9 : 1 water/methanol solutions.

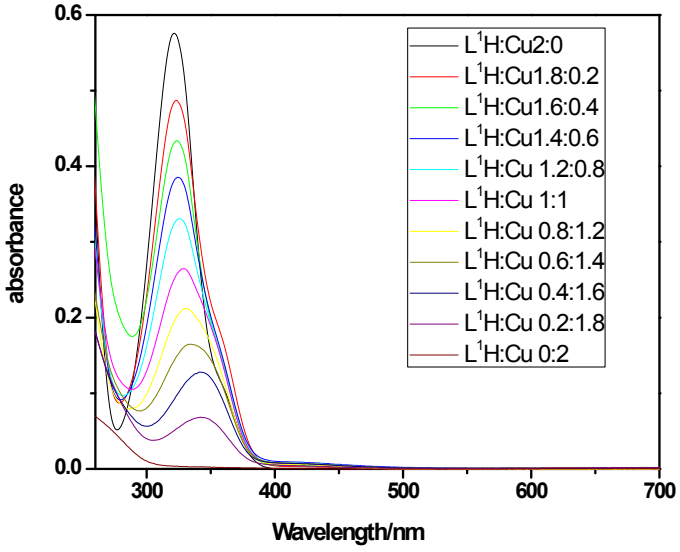


Figure SI7. Job's plot for **1OH** binding to Cu(II) (top) and to Fe(III) (bottom) in EtOH/H_2O 9:1; for the LMCT bands at 425 nm and 458 nm, respectively.

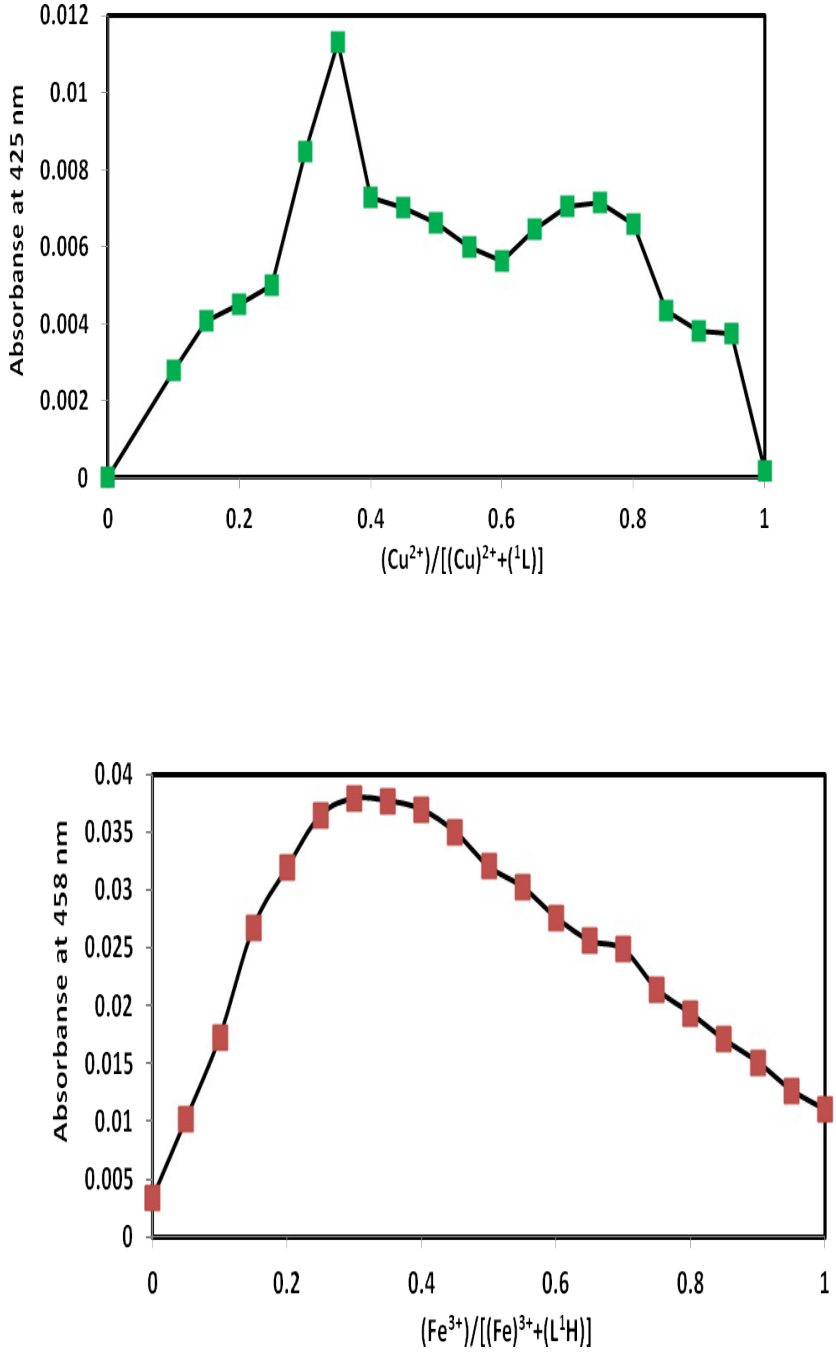


Figure SI8. Percentage of OH radical inhibition of **1OMe** (■) and **1OH** (•). Hydroxyl radicals were generated by photolysis of an aqueous solution containing

2.5 mM H_2O_2 and 10 mM DMPO using a 30 W, 365 nm UV-lamp. Each point represents the average of three independent experiments.

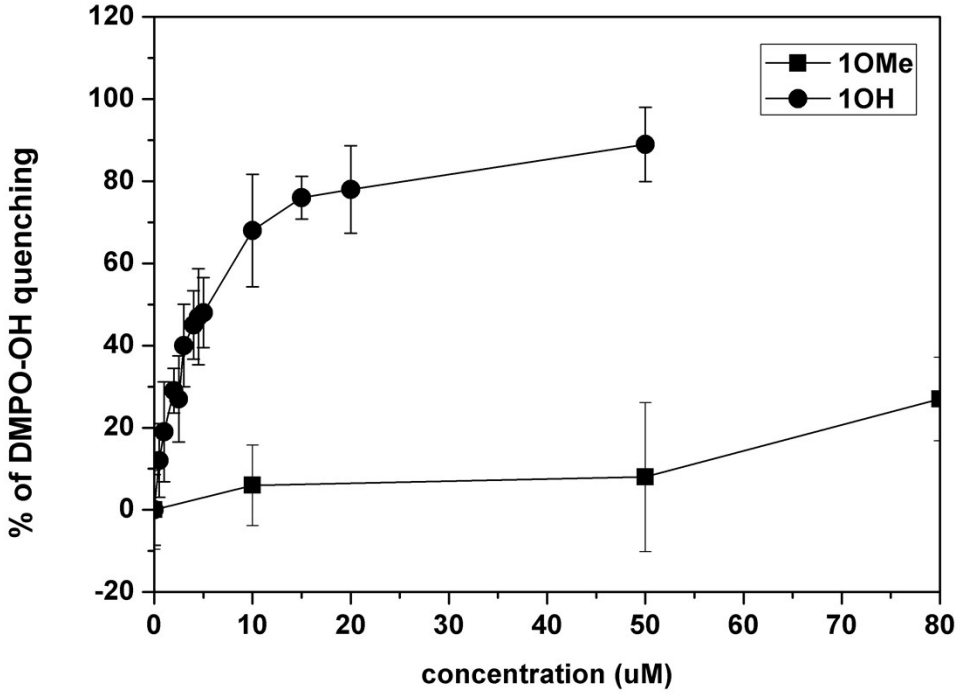


Figure SI9. Effect of test compounds on lipid peroxidation (MDA assay) on NSC-34 cells. Trolox, **1OH** and **1OMe** (100 μM) were added in DMSO (0.5 %

concentration) to the cultures. After 30 min of incubation, oxidative stress was induced by GO system for 1 h as described above (red bars) and MDA assay was conducted. Untreated by GO cells were used as a control (black bars). *, $p<0.05$, comparing to cells incubated with GO, MEAN ± SE, n=6.

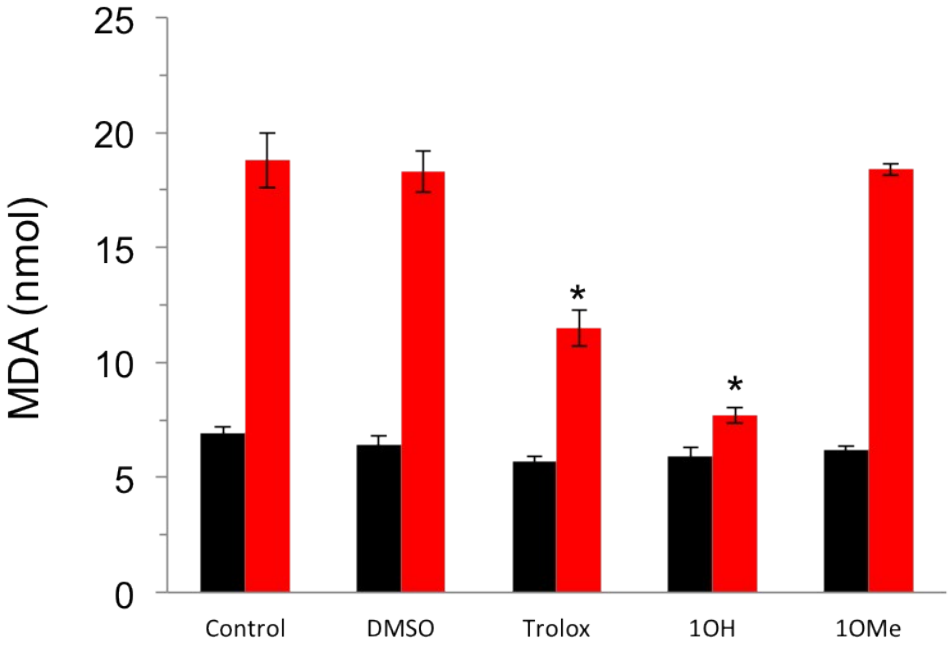


Figure SI10. Effect of **1OMe**, **1OH**, and Trolox on total antioxidant capacity as measured by the TEAC assay (see expt section). Conditions: cells: G93AhSOD1-NSC-34; concentrations: Trolox (1 mM), **1OH** (0.33 mM) and **1OMe** (0.33 mM); H_2O_2 (500 μM). After 30 mn incubation time, the cell lysates were submitted

TEAC assays. The $ABTS^{\bullet+}$ was monitored by UV/Vis measurements at λ = 750 nm. The results are expressed in TEAC units. *, $p<0.05$ in comparison with an absorption level of a sample contain DMSO only, **, $p<0.05$ in comparison with an absorption level of a treated by H_2O_2 cells.