

Electronic Supplementary Information for

**Nitric Oxide Release study of a Bio-inspired Copper(I)-Nitrito
Complex on Chemical and Biological Conditions**

Wan-Jung Chuang^a, Manmath Narwane^a, Hsing-Yin Chen^a, Chai-Lin Kao^{a,b}, Bin
Huang^c, Kuang-Mei Hsu^d, Yun-Ming Wang^{*,c,d}, Sodio C. N. Hsu^{*,a,b}

^a*Department of Medicinal and Applied Chemistry, Kaohsiung Medical University,
Kaohsiung 807, Taiwan*

^b*Department of Medical Research, Kaohsiung Medical University Hospital,
Kaohsiung 807, Taiwan*

^c*Department of Biomedical Science and Environmental Biology, Kaohsiung Medical
University, Kaohsiung 807, Taiwan*

^d*Department of Biological Science and Technology, Institute of Molecular Medicine
and Bioengineering, National Chiao Tung University, Hsinchu 300, Taiwan*

Contents

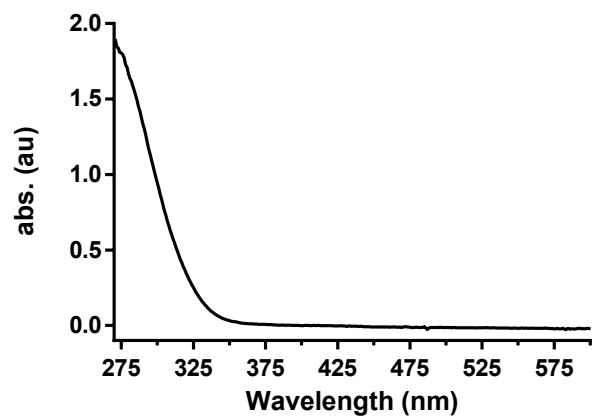
Fig. S1 Absorption spectra of complex 2(a) , 3(b) and 4(c) in CH ₂ Cl ₂ at room temperature.....	4
Fig. S2 FT-IR spectra of 2-(diphenylphosphino) aniline and complex 2 , 3 and 4 . (KBr pellet).	5
Fig. S3 FT-IR spectra for the ¹⁴ NO ₂ and ¹⁵ NO ₂ isotopomers of complex 4 (KBr pellet)	6
Fig. S4 ORTEP representation of complexes 2(a) and 3(b) (50% ellipsoids, hydrogen atoms not shown for clarity).....	7
Fig. S5 NO generation experiments for 4 . NO concentration performed by calibrating curve response with known concentrations of NO mixed with N ₂ (10 ppm, 20 ppm, 30 ppm and 40 ppm of NO in N ₂); molar quantities were calculated using the ideal gas equation.	8
Fig.S6 Time dependent detailed study of UV-spectrum observed in complex 4 after protonation with excess of TFA at 273 K.	8
Fig. S7. Eyring plot for the reactions of 1 and TFA.....	9
Fig. S8 Eyring plot for the reactions of 4 and TFA.....	9
Fig. S9 (a) Absorption spectral change of complex 3 for the titration of TFA (0~3 eq. TFA) in CH ₂ Cl ₂ at room temperature, Each spectrum was obtained after equilibration for 10 min. (b) Absorbance change at 300 nm over TFA concentration	10
Fig. S10 FT-IR spectra of complex 3 before and after protonation	11
Fig.S11 NMR titration spectra of complex 3 for the titration of TFA (0~2 eq. TFA) in CH ₂ Cl ₂ at room temperature, each spectra was obtained after equilibration for 10 min.	11
Fig. S12 (a) Absorption spectral change of complex 4 for the titration of TFA (0~3 eq. TFA) in CH ₂ Cl ₂ at room temperature, Each spectrum was obtained after equilibration for 10 min. (b) Absorbance change at 300 nm over TFA concentration.	12
Fig. S13 EPR spectra of complex 4 for the titration of TFA (0~1 eq. TFA) in CH ₂ Cl ₂ at room temperature, each spectra was obtained after equilibration for 10 mins.	13
Fig. S14 Fluorescence emission spectra of FA-OMe (1 μM) upon reaction with various amounts of DETA NONOate in 100 mM PBS buffer with 0.54% DMSO of pH 8.5 at 25.0 ± 0.1 °C(λ _{ex} 460 nm, λ _{em} 515 nm). Inset shows an enhancement in fluorescence intensity presents a linear correlation with concentration of DETA NONOate.	14

Fig. S15 Normalized fluorescence response of the FA-OMe (1 μ M) with DETA NONOate, **1**, and **4** (5 μ M) in different pH values (100 mM PBS buffer with 0.54% DMSO) and 25.0 ± 0.1 °C. Tabulated values indicates the NO release response shown by DETA NONOate compared with the complex **4** and complex **1**15

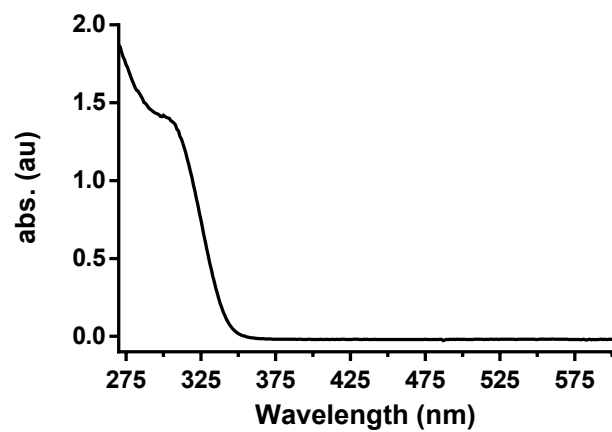
Tab. S1 First order rate constants for the reaction of **1** and **4** with excess TFA in CH_2Cl_2 at 273 K.15

Tab. S2 Selected Bond Lengths (\AA) and Angles ($^\circ$) for **2**, **3**, and **4**.16

(a)



(b)



(c)

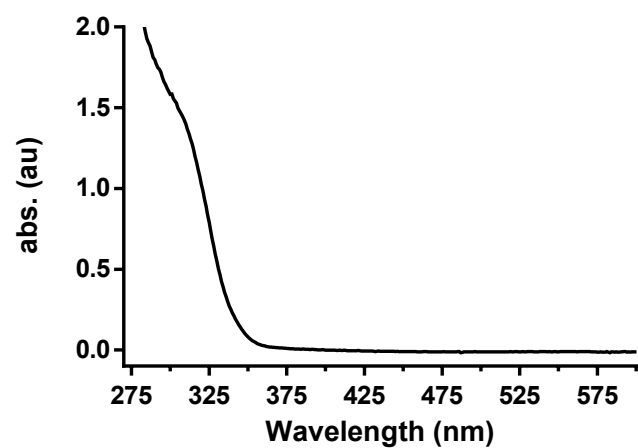


Fig. S1 Absorption spectra of complex **2(a)**, **3(b)** and **4(c)** in CH_2Cl_2 at room temperature.

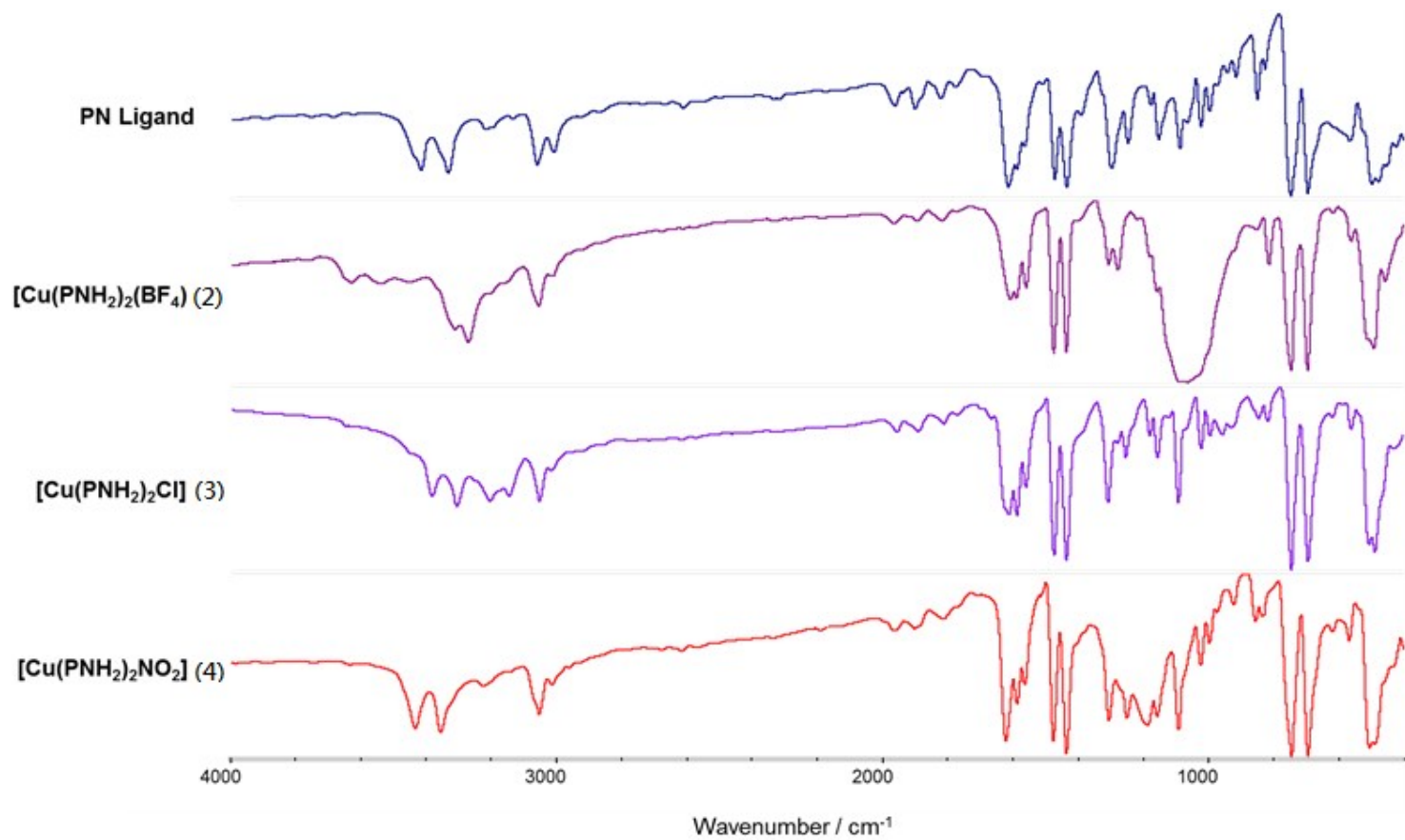


Fig. S2 FT-IR spectra of 2-(diphenylphosphino) aniline and complex **2**, **3** and **4**. (KBr pellet).

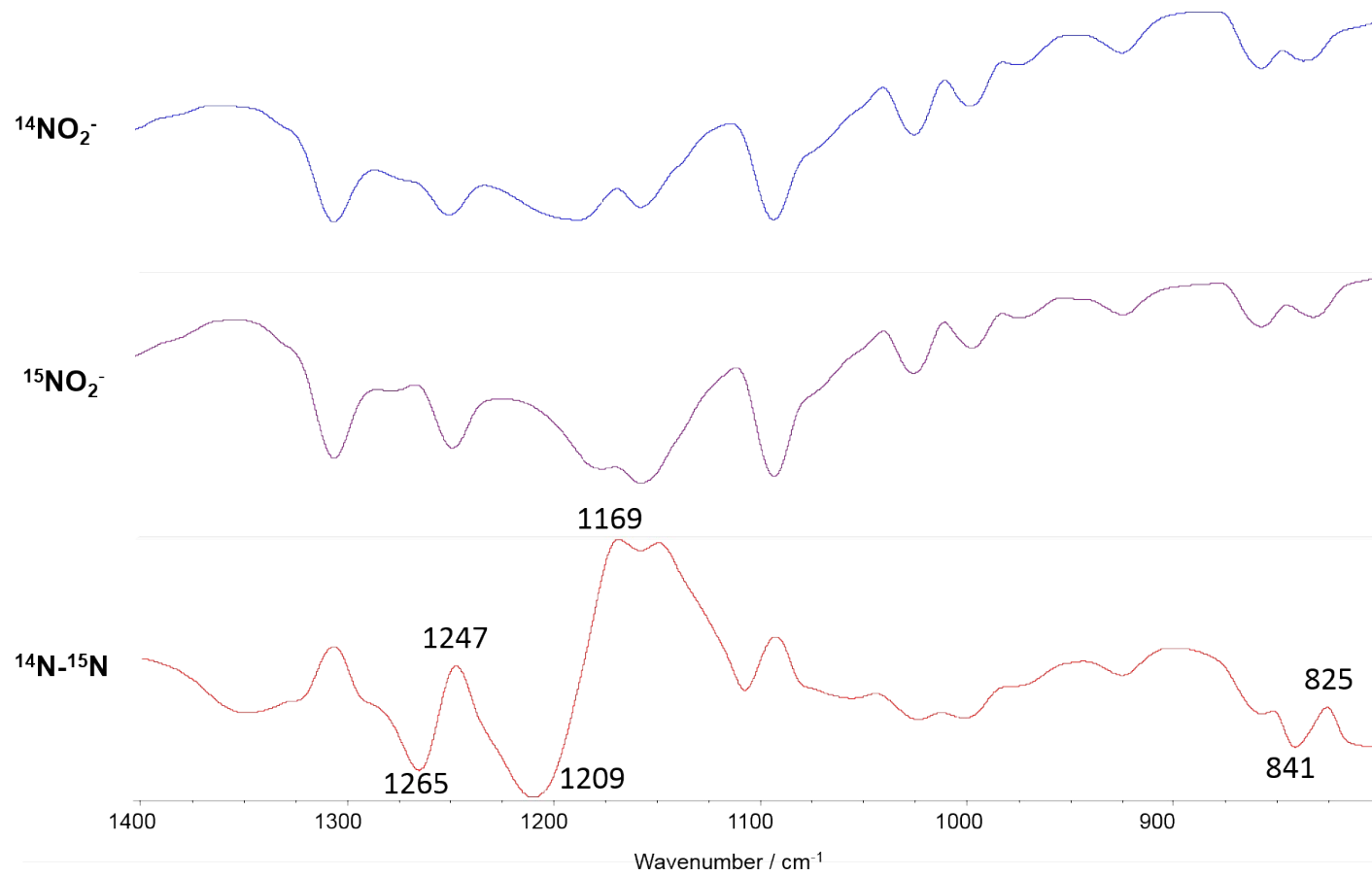
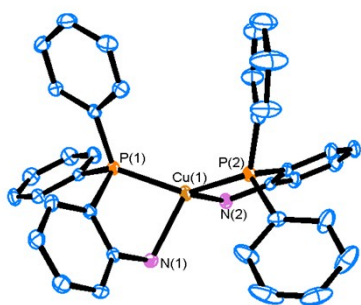
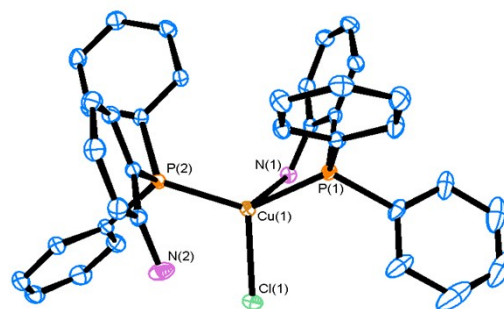


Fig. S3 FT-IR spectra for the ¹⁴NO₂ and ¹⁵NO₂ isotopomers of complex 4 (KBr pellet)



(a)



(b)

Fig. S4 ORTEP representation of complexes **2(a)** and **3(b)** (50% ellipsoids, hydrogen atoms not shown for clarity).

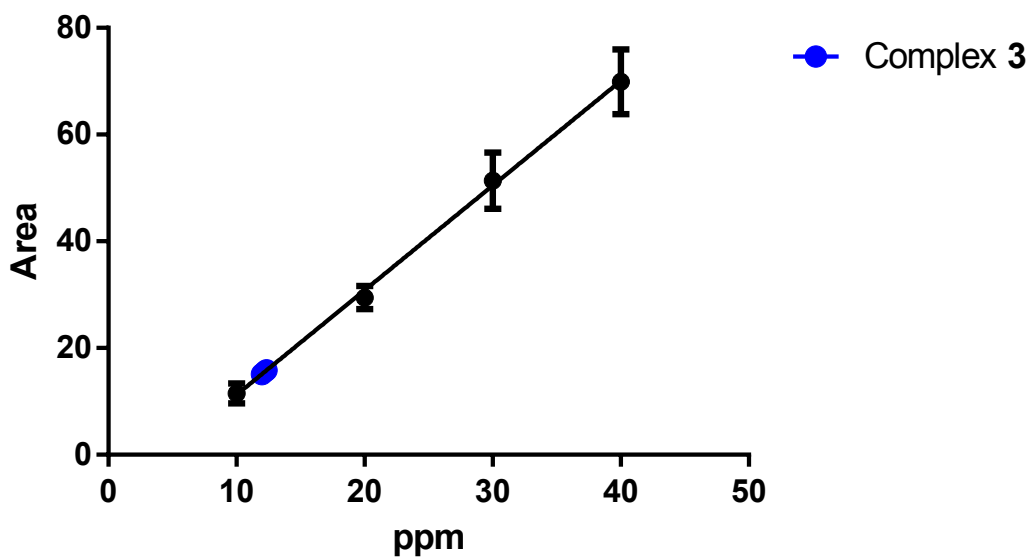


Fig. S5 NO generation experiments for **4**. NO concentration performed by calibrating curve response with known concentrations of NO mixed with N₂ (10 ppm, 20 ppm, 30 ppm and 40 ppm of NO in N₂); molar quantities were calculated using the ideal gas equation.

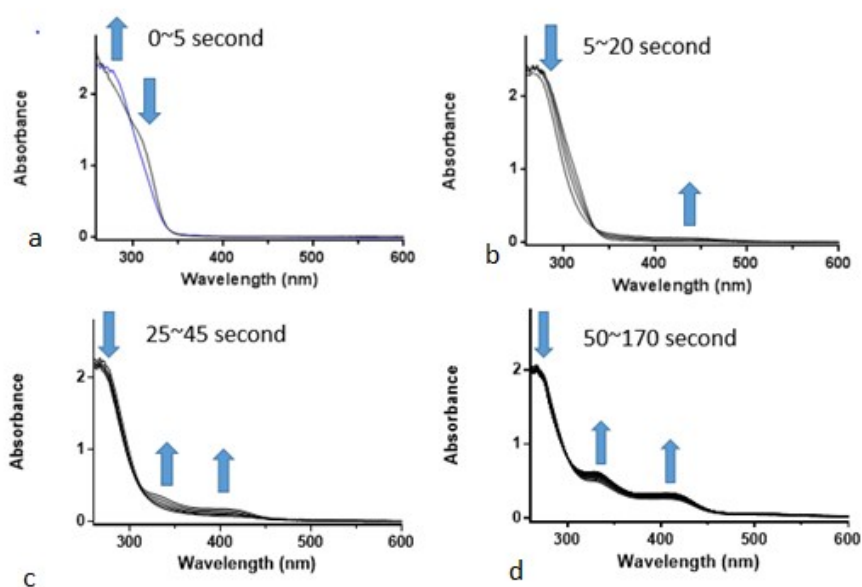


Fig.S6 Time dependent detailed study of UV-spectrum observed in complex **4** after protonation with excess of TFA at 273 K.

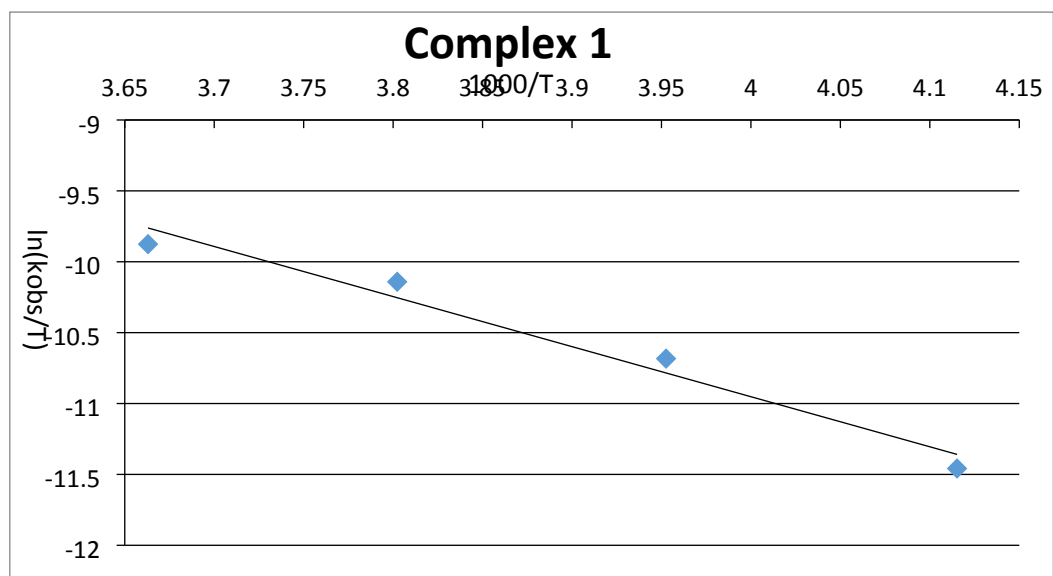


Fig. S7. Eyring plot for the reactions of **1** and TFA.

The ΔH^\ddagger and ΔS^\ddagger values are determined from the Eyring equation, $k' = (kT/h)\exp(\Delta S^\ddagger/R) \exp(-\Delta H^\ddagger/RT)$.

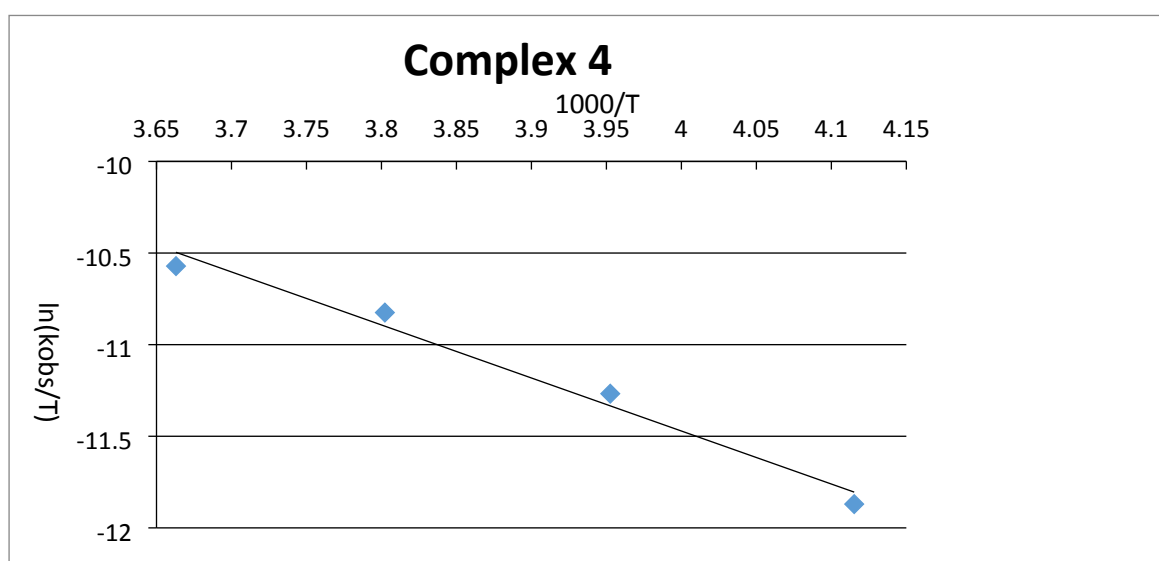
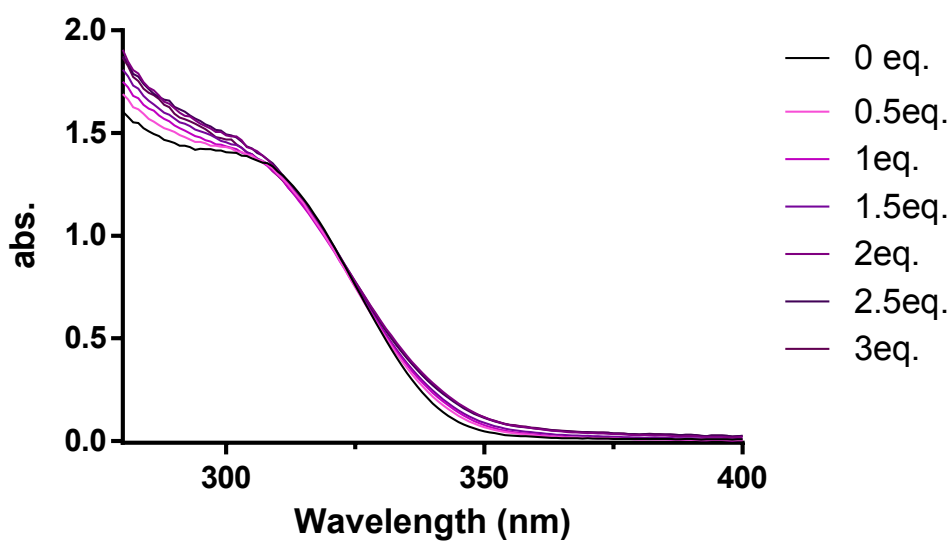


Fig. S8 Eyring plot for the reactions of **4** and TFA.

The ΔH^\ddagger and ΔS^\ddagger values are determined from the Eyring equation, $k' = (kT/h)\exp(\Delta S^\ddagger/R) \exp(-\Delta H^\ddagger/RT)$.

(a)



(b)

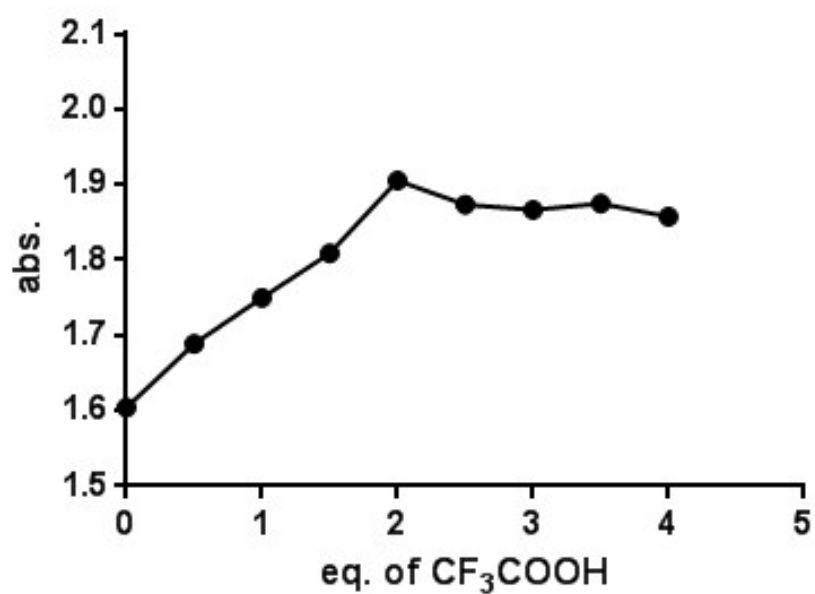


Fig. S9 (a) Absorption spectral change of complex **3** for the titration of TFA (0~3 eq. TFA) in CH₂Cl₂ at room temperature, Each spectrum was obtained after equilibration for 10 min. (b) Absorbance change at 300 nm over TFA concentration

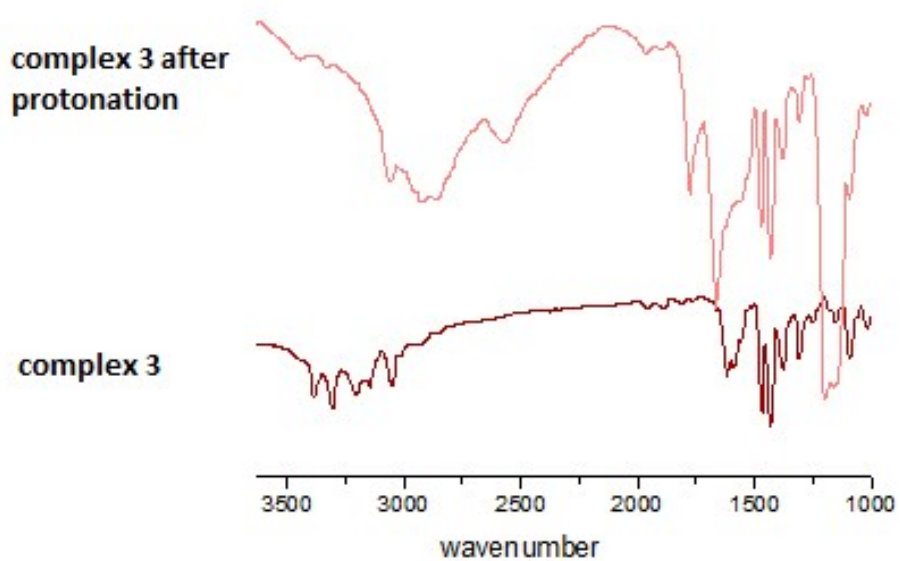


Fig. S10 FT-IR spectra of complex **3** before and after protonation

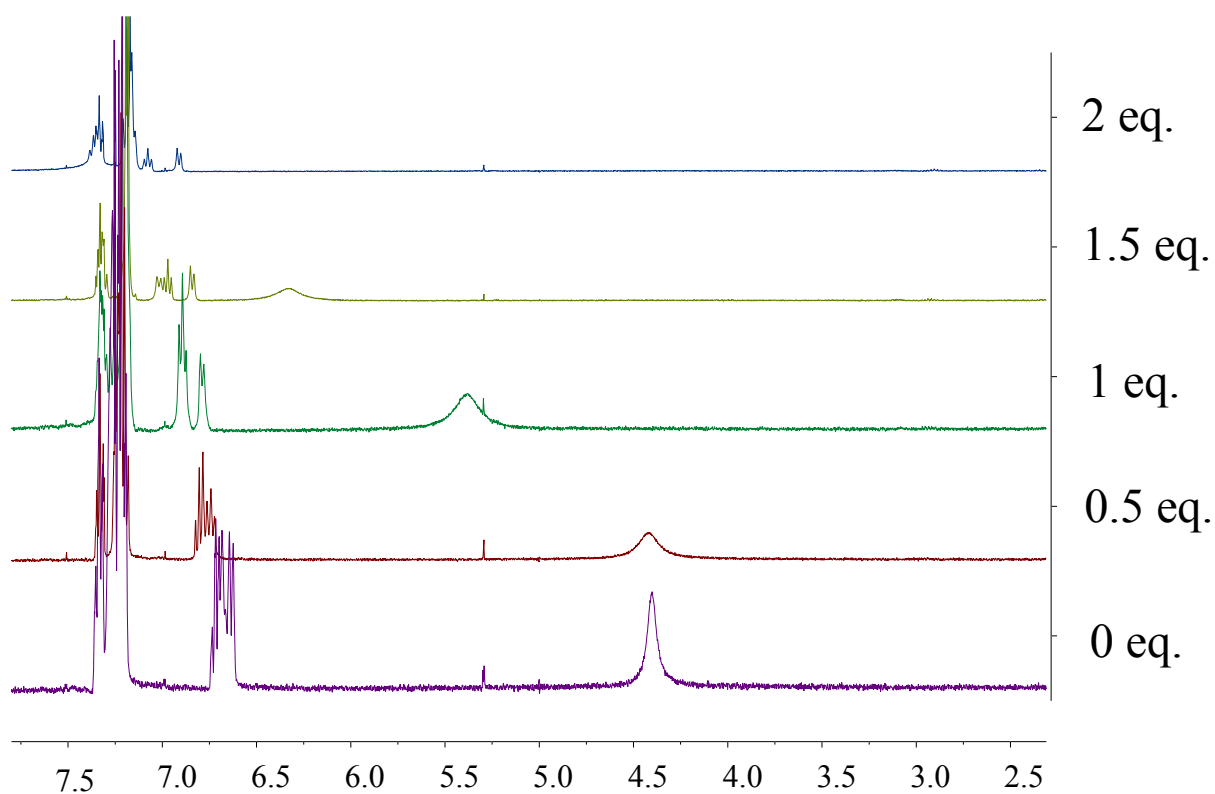
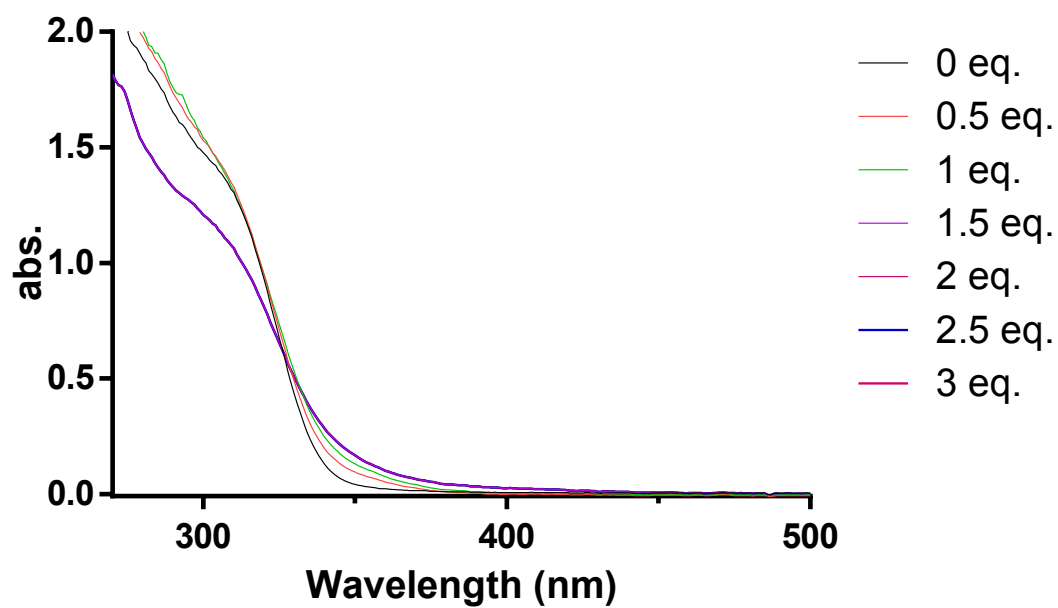


Fig.S11 NMR titration spectra of complex **3** for the titration of TFA (0~2 eq. TFA) in CH₂Cl₂ at room temperature, each spectra was obtained after equilibration for 10 min.

(a)



(b)

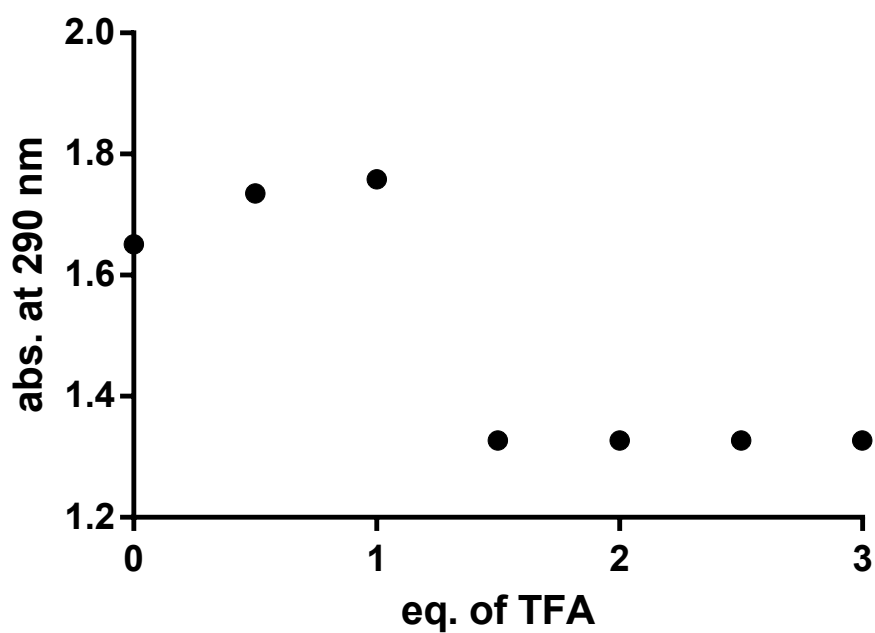


Fig. S12 (a) Absorption spectral change of complex **4** for the titration of TFA (0~3 eq. TFA) in CH_2Cl_2 at room temperature, Each spectrum was obtained after equilibration for 10 min. (b) Absorbance change at 300 nm over TFA concentration.

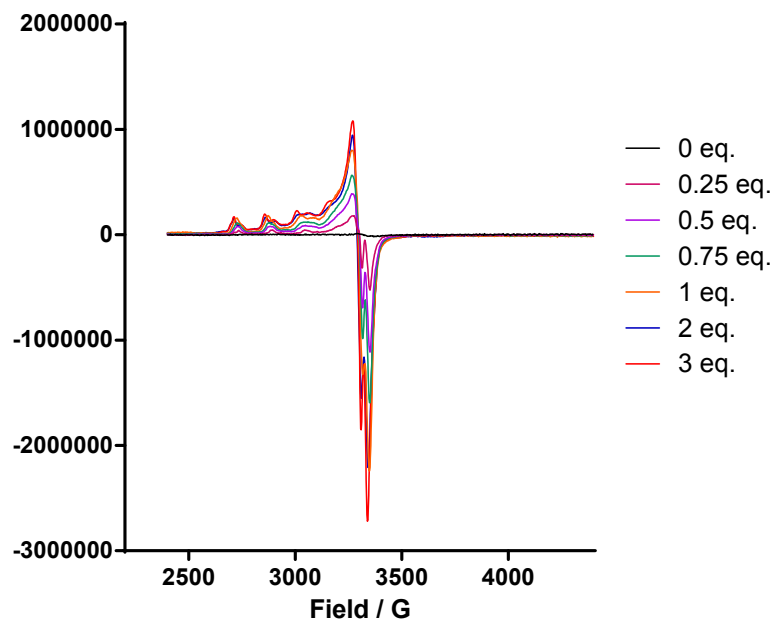


Fig. S13 EPR spectra of complex **4** for the titration of TFA (0~1 eq. TFA) in CH₂Cl₂ at room temperature, each spectra was obtained after equilibration for 10 mins.

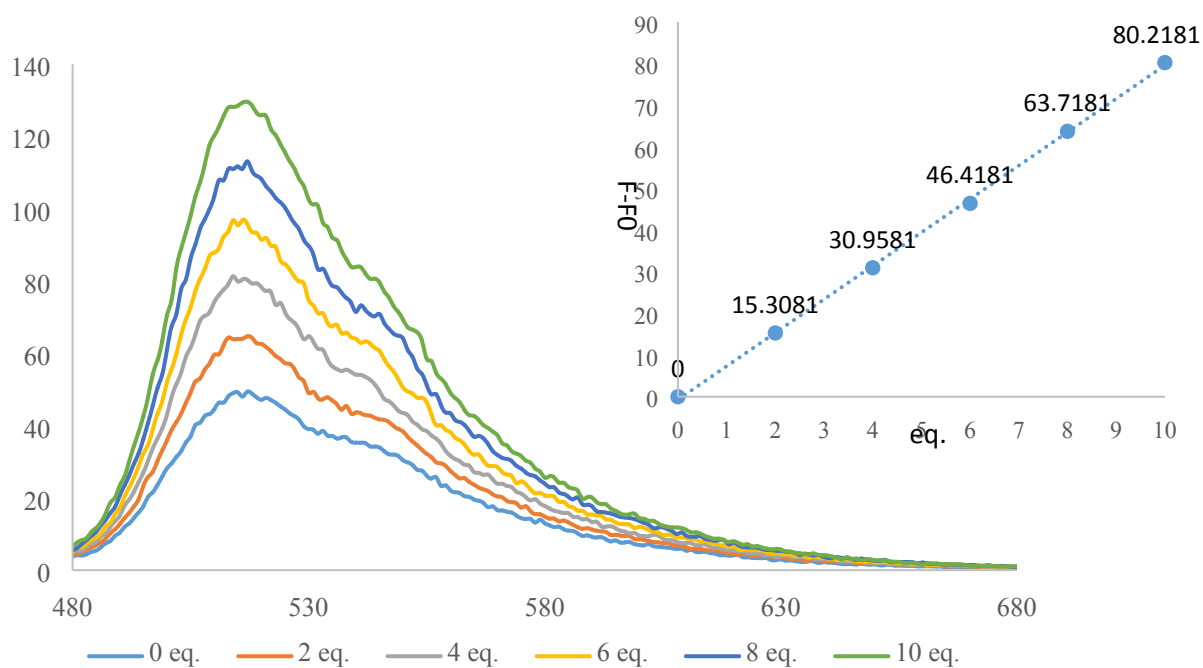


Fig. S14 Fluorescence emission spectra of FA-OMe (1 μ M) upon reaction with various amounts of DETA NONOate in 100 mM PBS buffer with 0.54% DMSO of pH 8.5 at 25.0 ± 0.1 $^{\circ}$ C (λ_{ex} 460 nm, λ_{em} 515 nm). Inset shows an enhancement in fluorescence intensity presents a linear correlation with concentration of DETA NONOate.

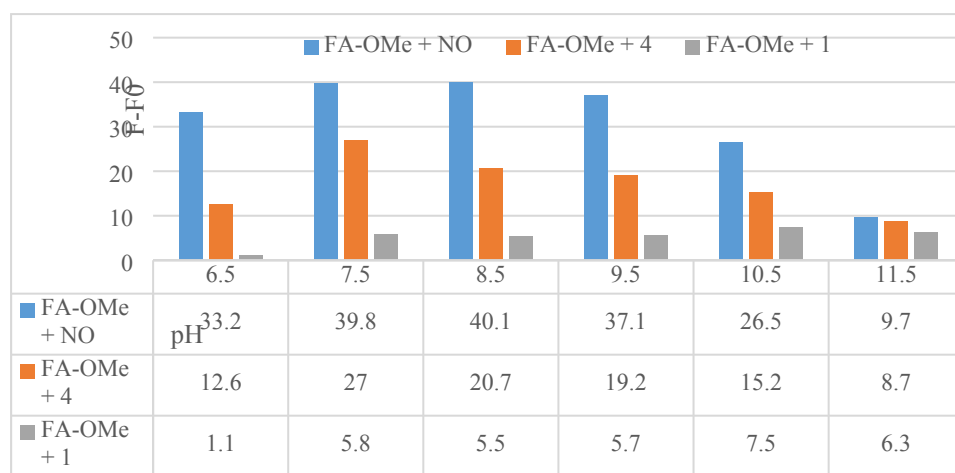


Fig. S15 Normalized fluorescence response of the FA-OMe (1 μM) with DETA NONOate, **1**, and **4** (5 μM) in different pH values (100 mM PBS buffer with 0.54% DMSO) and 25.0 ± 0.1 $^{\circ}\text{C}$. Tabulated values indicates the NO release response shown by DETA NONOate compared with the complex **4** and complex **1**.

Tab. S1 First order rate constants for the reaction of **1** and **4** with excess TFA in CH_2Cl_2 at 273 K.

[TFA] / mM	$k_{\text{obs}}/\text{s}^{-1}$ (complex 1)	$k_{\text{obs}}/\text{s}^{-1}$ (complex 4)
9.41	$(1.4 \pm 0.04) \times 10^{-2}$	$(6.9 \pm 0.62) \times 10^{-3}$
18.82	$(2.1 \pm 0.02) \times 10^{-2}$	$(1.2 \pm 0.04) \times 10^{-2}$
28.23	$(2.9 \pm 0.02) \times 10^{-2}$	$(1.9 \pm 0.05) \times 10^{-2}$
37.64	$(4.0 \pm 0.05) \times 10^{-2}$	$(2.2 \pm 0.22) \times 10^{-2}$
47.05	$(4.9 \pm 0.01) \times 10^{-2}$	$(3.0 \pm 0.44) \times 10^{-2}$

^a Average values of three experimental values.

Tab. S2 Selected Bond Lengths (Å) and Angles (°) for **2**, **3**, and **4**.

	2	3	4
Cu(1)-P(1)	2.230(1)	2.2781(7)	2.233(1)
Cu(1)-P(2)	2.225(1)	2.2410(6)	2.243(1)
Cu(1)-N(1)	2.212(3)	2.251(2)	-
Cu(1)-N(2)	2.071(3)	-	-
Cu(1)-O(2)	-	-	2.257(3)
Cu(1)-O(1)	-	-	2.226(3)
Cu(1)-halide	-	2.3073(8)	-
P(1)-Cu(1)-P(2)	131.66(4)	126.05(3)	129.58(4)