Supporting Information

A Computer-Designed Macrocyclic Zinc Receptor

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(Experimental procedures, Cartesian coordinates for **2**, and Job plots.)

General. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker AC-250 (250 MHz ^{1}H) or Varian Gemini-2300 (300 MHz ^{1}H, 75 MHz ^{13}C) spectrometer. ^{1}H spectra were referenced to the residual proton peak of the deuterated solvent. ^{13}C spectra were referenced to $CDCl_3$ (77.00 ppm). Chemicals were purchased from Aldrich and used as received. Novozym-435 was obtained from Novozymes. AM1 calculations were performed using PC Spartan Pro from Wavefunction, Inc. Triflate salts of Ni(II) and Co(II) were prepared from the carbonate salts as described previously.[S1]

8a, 8b-Dihydro-as-indacene 7

N,N-dimethylaminofulvene **5** (1 g, 8.4 mmol) was dissolved in dry THF(50 mL). The flask was flushed with N_2. Lithium wire (1 g) was cut and added with naphthalene (54 mg, 0.42 mg) into the flask. The reaction was stirred at room temperature in the dark for 8h. The mixture was filtered and the filtrate was evaporated. The crude product was purified by chromatography with hexanes to give **7** (0.42 g, 2.7 mmol, 32%). ^{1}H NMR($CDCl_3$) δ: 6.5-7.0 (m, 6H), 2.52(s, 2H); ^{13}C NMR($CDCl_3$) δ: 148.7, 136.9, 134.8, 125.2, 122.5, 56.9;. MS (EI) Calcd for $C_{12}H_{10}$ 154.2, found m/z: 153 (M^{+}-H). The ^{1}H NMR spectrum was consistent with a published spectrum.[20] An approximately 3:1 mixture of 1,8-dihydro-as-indacene and 1,6-dihydro-as-indacene (0.08 g) was also isolated.

1, 8-Dihydro-as-indacene 8

A solution of **7** (0.4 g, 2.6 mmol) in cyclohexane (50 mL) was heated for 3h at reflux. The solvent was evaporated to give **8** (0.4 g, 2.6 mmol), which was used in the next step without further purification. ^{1}H NMR ($CDCl_3$) δ: 7.32 (s, 2H), 6.93 (dt, 2H, J_1=3Hz, J_2=1Hz), 6.49 (dt, 2H, J_1=3Hz, J_2=1Hz), 3.42 (t, 2H, J=3Hz). ^{13}C NMR ($CDCl_3$) δ: 136.0, 134.7, 128.4, 123.0, 121.7, 24.9. The ^{1}H NMR spectrum was consistent with a published spectrum.[20]

1,2,3,6,7,8-Hexahydro-as-indacene-2,7-diol 9

A suspension of (+)-diisopinocampheylborane (2.60 mmol) in diglyme (15 mL) was prepared as described.[22] 1,8-Dihydro-as-indacene **8** (191 mg, 1.24 mmol) was added and the reaction was stirred at 0°C for 3 hours. The ice bath was then removed and stirring was continued at room temperature for 12 hours. Water (10 mL) was added followed by dropwise addition of aq. hydrogen peroxide (1.5 mL) and 30% aq. NaOH (2.5 mL). The mixture was heated to 40°C for 50 min, water (30 mL) was added and the mixture was extracted with diethyl ether (4 × 30mL). The organic layer was dried over magnesium sulfate, concentrated by rotary evaporation, and further dried under high vacuum. The crude product was purified by column chromatography on silica gel eluted with 1:4 hexanes/ethyl acetate to give **9** containing stereoisomeric impurities (151mg, 0.79 mmol, 64%). ^{1}H NMR ($CDCl_3$) δ: 7.10 (s, 2H), 4.40 (m, 2H), 3.22 (m, 4H), 3.04 (m, 4H), 1.94 (broad, 2H); ^{13}C NMR ($CDCl_3$) δ:139.4, 137.1, 123.2, (73.38, 73.36), (42.57, 42.53), (41.01, 40.98), double peaks due to the presence of isomers are shown in parenthesis; MS(EI) Calc. for $C_{12}H_{14}O_2$, 190.2, found m/z: 190(M^+).

MTPA esters of 1,2,3,6,7,8-hexahydro-as-indacene-2, 7-diol

To a solution of **9** (40mg, 0.21mmol) in THF (20 mL) was added (S)-(-)-α-methoxy-α-(trifluoromethyl)phenylacetic acid (150mg, 0.63mmol), dicyclohexylcarbodiimide (86mg, 0.42mmol), and dimethylaminopyridine (51mg, 0.42mmol). The reaction was stirred for 20 h at room temperature. The mixture was diluted with dichloromethane (60 mL) and washed with 1M HCl (30mL), saturated $NaHCO_3$ solution (30mL) and brine (30 mL). The organic layer was dried over magnesium sulfate and

the solvent was removed under reduced pressure. The crude product was purified by column chromatography on silica gel with 20:1 hexanes/ethyl acetate to give the Mosher diester mixture. The mixture was further purified by preparative TLC on silica gel eluted with 10:1 hexanes/ethyl acetate. The upper band contained the Mosher diester assigned to (2S, 7S)-1,2,3,6,7,8-hexahydro-as-indacene-2, 7-diol. ^{1}H NMR ($CDCl_3$) δ: 7.49-7.40 (m, 10H), 7.06 (s, 2H), 5.82 (m, 2H), 3.89-2.85 (m, 24H); ^{19}F NMR ($CDCl_3$), δ: -71.93. The lower band was assigned to the Mosher diester of (2S, 7R)-1,2,3,6,7,8-hexahydro-as-indacene-2, 7-diol. ^{1}H NMR ($CDCl_3$) δ: 7.49-7.40 (m, 10H), 7.06 (s, 2H), 5.82 (m, 2H), 3.89-2.85 (m, 24H); ^{19}F NMR ($CDCl_3$), δ: - 71.97, -71.99.

Stereochemical enrichment of (2S,7S)-1,2,3,6,7,8-hexahydro-as-indacene-2, 7-diol 9

To a solution of **9** (60mg, 0.32mmol) in dichloromethane (10 mL) was added Novozym-435 (30mg) and the suspension was stirred vigorously at room temperature. A solution of vinyl acetate (60mg, 0.32mmol) in dichloromethane (2 mL) was added dropwise. The reaction was monitored by ^{1}H NMR analysis of removed samples until 30% acylation of hydroxyl groups was reached. The suspension was filtered and the filtrate was evaporated under reduced pressure. The crude product was purified by chromatography eluted by 1:4 hexanes/ethyl acetate to give (2S, 7S)-1,2,3,6,7,8-hexahydro-as-indacene-2, 7-diol **9** (24mg, 0.13 mmol, 40%). $[\alpha]^{20}_{D}$=7.1°. ^{1}H NMR ($CDCl_3$) δ: 7.10 (s, 2H), 4.40 (m, 2H), 3.22 (m, 4H), 3.04 (m, 4H), 1.94 (broad, 2H); ^{13}C NMR ($CDCl_3$) δ: 139.3, 137.0, 123.0, 73.1, 42.2, 40.7. The product was converted to Mosher ester according to the above procedure. ^{19}F NMR($CDCl_3$): δ -71.92.

(2R, 7R)-2,7-Diazido-1,2,3,6,7,8-hexahydro-as-indacene 12

To a solution of (2S, 7S)-1,2,3,6,7,8-hexahydro-as-indacene-2,7-diol **9** (123mg, 65mmol) and triethylamine (0.25mL, 1.82mmol) in dichloromethane (20mL) at 0°C was added methanesulfonyl chloride (0.11mL, 1.43mmol) dropwise over 10 min. After 2h at 0°C, the reaction mixture was diluted with dichloromethane (50 mL) and washed with aq. HCl (1M, 30mL), saturated aq. $NaHCO_3$ (30mL) and brine (30mL). The organic layer was dried over magnesium sulfate and the solvent was removed

under reduced pressure to give the mesylate ester. The mesylate ester was dissolved in anhydrous DMSO (20 mL), sodium azide (687mg, 3.25mmol) was added and the reaction was heated to 50° for 6h. The reaction was diluted with ethyl acetate (200 mL) and washed with water (2×100mL). The organic layer was dried over magnesium sulfate and concentrated under reduced pressure. The product was purified by column chromatography on silica gel eluted with 10:1 hexanes/ethyl acetate to give **12** (97mg, 0.40 mmol, 63% from **9**). ^{1}H NMR ($CDCl_3$) δ: 7.09 (s, 2H), 4.40 (m, 2H), 3.19 (m, 4H), 2.94 (m, 4H). ^{13}C NMR ($CDCl_3$) δ: 139.1, 136.2, 123.2, 62.0, 38.8, 37.4. MS (EI) Calc. for $C_{12}H_{12}N_6$ 240.2, found m/z 184(M^+-$2N_2$).

(2R, 7R)-N,N'-Dimethyl-1,2,3,6,7,8-hexahydro-as-indacene-2,7-diamine 13

To a solution of **12** (97mg, 0.40mmol) in dichloromethane (10mL) was added dimethylbromoborane (0.39mL, 4mmol) dropwise. The reaction was stirred overnight at room temp. and the reaction was quenched with methanol (5 mL). The mixture was diluted with dichloromethane (20 mL) and washed with aq. HCl (1M, 3×10mL). The aqueous layers were combined, placed in an ice water bath and solid sodium hydroxide (4.8 g) was added. The resulting solution was extracted with dichloromethane (3×30mL). The organic layers were combined and dried over magnesium sulfate. The solvent was removed under reduced pressure to give **13** (67mg, 0.31 mmol, 78%). ^{1}H NMR ($CDCl_3$), δ: 7.00 (s, 2H), 3.54 (m, 2H), 3.09 (m, 2H), 2.48 (s, 6H). ^{13}C NMR ($CDCl_3$): 139.9, 137.8, 122.6, 61.5, 39.5, 38.0, 34.7. MS (EI) Calc. for $C_{12}H_{20}N_2$ 216.3, found m/z 216 (M^+).

5-Ethoxycarbonyl-5'-carboxyl-2,2'-bipyridine

5,5'-Bis(ethoxycarbonyl)-2,2'-bipyridine (2.72g, 10mmol) was made in two steps from 5,5'-dimethyl-2,2'-bipyridine as previously described.[27] The starting material was dissolved in 2:1 THF/ethanol (150 mL) and a solution of KOH (0.50g, 9mmol) in THF/ethanol (30mL) was added dropwise over 1h. The resulting suspension was stirred at room temperature for 3h and the solvent was evaporated. The residue was dissolved in 50mL water and the solution was extracted with chloroform (2 x 20 mL). The aqueous phase was acidified to pH 4 and the resulting precipitate was filtered and

dried in air to yield 5-ethoxycarbonyl-5'-carboxyl-2,2'-bipyridine (2.28g, 8.38 mmol, 84%). ^{1}H NMR (DMSO- d_6) δ: 9.24 (s, 2H), 8.61 (m, 2H), 8.51 (m, 2H), 4.43 (q, 2H, J=6Hz), 1.41 (t, 3H, J=6Hz); ^{13}C NMR (DMSO-d_6) δ: 165.9, 164.4, 157.5, 157.1, 150.3, 150.0, 138.4, 138.2, 121.1, 61.3, 14.1.

Compound 15

5-Ethoxycarbonyl-5'-carboxyl-2,2'-bipyridine (100mg, 0.37mmol) was dissolved in thionyl chloride (2 mL) and the reaction was stirred at reflux for 4h. The solvent was removed by evaporation under reduced pressure to give the acid chloride as a white solid. The acid chloride was also dissolved in dichloromethane (5mL) and the resulting solution was added dropwise into a solution of (2R, 7R)-N,N'-dimethyl-1,2,3,6,7,8-hexahydro-as-indacene-2,7-diamine (29mg, 0.13mmol) and triethylamine (37μL, 0.37mmol) in dichloromethane (10mL) over 10 min at 0°C. The ice bath was removed and the reaction was stirred at room temperature overnight. The mixture was diluted with dichloromethane (30 mL) and washed with saturated aq. sodium bicarbonate solution (20mL) and brine (20 mL). The organic layer was dried over magnesium sulfate and evaporated under reduced pressure. The product was purified by column chromatography on silica gel eluted with 20:1 chloroform/methanol to give **15** (73mg, 0.10 mmol, 77%). ^{1}H NMR ($CDCl_3$, 20º C) δ: 9.27 (d, 2H, J=1.5Hz), 8.78 (d, 2H, J=1.5Hz), 8.42-8.56 (m, 6H), 7.95 (dd, 2H, J_1=6Hz, J_2=1.5Hz), 7.03 (broad, 2H), 4.8-5.0 (broad, 1H), 5.4-6.0 (broad, 1H), 4.43 (q, 4H, J=6Hz), 2.6-3.6 (m, 14H), 1.41 (t, 6H, J=7Hz). At 50º C the signal at 7.03 became a sharp singlet while the broad signals at 4.8-5.0 and 5.4-6.0 coalesced into a single broad peak centered at about 5.0 ppm. MS (FB^+) Calc. for $C_{42}H_{40}N_6O_6$ 724.8 , found m/z 725.5.

Diacid of 15

A solution of **15** (73mg, 0.10 mmol) in a mixed solvent of aq. NaOH (1M, 1mL, 0.1mol), THF (10mL) and methanol (10mL) was stirred at room temperature overnight. The solvent was removed under reduced pressure and the residue was dissolved in water (10 mL). Aq. HCl (1M) was added dropwise until the pH was 4. The suspension was centrifuged and the clear solution was removed. The resulting residue was dried over an air stream to give the diacid (65mg, 0.10 mmol, 97%). ^{1}H NMR (d_6-

MDSO), δ: 9.33 (d, 2H, J=1.5Hz), 8.82 (d, 2H, J=1.5Hz), 8.47-8.61 (m, 6H), 7.85 (dd, 2H, J_1=6Hz, J_2=1.5Hz), 7.10 (broad, 2H), 4.8-5.0 (broad, 1H), 5.4-6.0 (broad, 1H), 2.6-3.6 (m, 14H).

Cyclic receptor 4

To a solution of the diacid above (78mg, 0.12mmol), and **13** (26mg, 0.12mmol), in dimethylformamide (25 mL) was added 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (EDC) (46mg, 0.24mmol) and HOBT (32mg, 0.24mmol). The reaction was stirred at room temperature for 24h and the solvent was removed under high vacuum. The residue was dissolved in chloroform (40 mL) and washed with water (20mL), saturated aq. sodium bicarbonate solution (20mL) and brine (20mL). The organic layer was dried over magnesium sulfate and the solvent was removed under reduced pressure. The product was purified by column chromatography on silica gel with ethyl acetate followed by 50:1 chloroform/methanol to give **4** (25mg, 0.03 mmol, 25%). ^{1}H NMR ($CDCl_3$), δ: 8.64 (m, 4H), 8.42 (m, 4H), 7.85 (m, 4H), 7.08 (m, 4H), 4.2-6.0 (broad, 4H), 2.5-3.5 (m, 28H). MS(FB^+) Calc. for $C_{52}H_{48}N_8O_4$ 849.0, found m/z 849.4.

Calorimetry experiments

All titrations were performed on a Calorimetry Sciences Corporation 4200 isothermal titration calorimeter at 25º C. The 1.3 mL receptor **4** solutions were titrated with 250-μL ligand solutions in 10μL injections. The concentrations of receptor and ligand solutions respectively in acetonitrile were 0.23 mM and 2.15 mM for $Zn(OTf)_2$, and 0.20 mM and 6.95 mM for $Cu(OTf)_2$. The titration curve was fit and the binding constant calculated using BindWorks 3.0 software based on the following equation.

$$Q = V \cdot \Delta H \cdot \left[[M]_t + \frac{1 + [R]_t\, n\, K_a + \sqrt{(1 + [R]_t\, n\, Ka - [M]_t\, K_a)^2 + 4\, K_a\, [M]_t}}{2K_a} \right]$$

Q: cumulative heat

V: volume of cell

ΔH: enthalpy of binding

K_a: binding constant

n: number of binding sites

$[M]_t$: total metal ion concentration

$[R]_t$: total receptor concentration

Fluorescence experiments

The fluorescence titrations were performed with an Aminco-Bowman Series 2 Spectrometer at 25º C. To 3.0 mL solutions of **4** (1.00×10^{-5} M) in acetonitrile were added the ligand solution from a 100-μL syringe. 20 injections of 10μL each of the metal triflate solution (6.00×10^{-4} M in acetonitrile) were added successively. The fluorescence emissions were scanned from 310 to 380 nm at the excitation wavelength of 290 nm. Binding constants were determined from fluorescence intensities at 358 nm. The relative fluorescence was plotted against free metal ion concentration. The curve was fit and the binding constants were calculated in Origin 7.0.

For 1:1 binding, the relative fluorescence is expressed as:

$$\frac{F - F_0}{F_{max} - F_0} = \frac{[M]}{[M] + K_d}$$

F: total fluorescence intensity

F_0: receptor fluorescence intensity before titration

F_{max}: total fluorescence intensity at saturation

[M]: free metal ion concentration

K_d: dissociation constant

[M] was calculated from

$$[M] = [M]_t - [R]_t \frac{F - F_0}{F_{max} - F_0}$$

$[R]_t$: total receptor concentration

$[M]_t$: total metal ion concentration

For 1:2 binding the relative fluorescence is expressed as:

$$\frac{F - F_0}{F_{max} - F_0} = \frac{[M]^2 + K_d[M]/2}{[M]^2 + K_d[M] + K_d^2}$$

[M] was calculated from

$$[M] = [M]_t - 2[R]_t \frac{F - F_0}{F_{max} - F_0}$$

These equations are based on the assumptions that the two binding sites are independent of each other and that the 1:1 complex gives half the fluorescence intensity increase of the 1:2 complex.

Job plots were obtained by measuring fluorescence intensity with changing mole fraction of the metal triflate.[S2,S3] Total concentration of metal triflate plus receptor **4** was kept constant at 5.00×10^{-5} M and the mole fraction of metal triflates was varied from 0 to 1. The relative fluorescence intensity was plotted as the difference between the observed fluorescence intensity and the fluorescence intensity of the same concentration of free receptor.

References

S1 M. T. Jansky and J. T. Yoke, *J. Inorg. Nucl. Chem.*, 1979, **41**, 1707.

S2 J. Zhou and T. D. James, *Chem. Commun.*, 2005, 1889.

S3 D. J. Oh, M. S. Han and D. H. Kim, *Bull. Korean Chem. Soc.*, 2004, **25**, 1495.

Table S1. Cartesian coordinates for **2**.

Zn	0.000770588	-0.000763020	0.001248276
N	-1.793499971	1.022581635	0.307777916
C	-4.325552700	2.218343481	0.227162245
C	-2.385482660	1.304632976	-0.903230684
C	-2.438758137	1.340860803	1.444482868
C	-3.718267746	1.953118434	1.463367025
C	-3.658645744	1.898692844	-0.959455422
H	-1.927622845	1.097420069	2.399829548
H	-4.146597422	2.111006911	-1.923675131
H	-5.328406564	2.680822482	0.190685704
N	-0.462438520	0.247482483	-2.019750073
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C	0.224864082	-0.091675893	-3.125395616
C	-1.636704614	0.955249577	-2.148419778
C	-2.108147927	1.340396662	-3.415912784
C	-0.196831671	0.251997210	-4.435482290
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N	0.400049821	-1.826868814	0.932225297
C	1.256264134	-3.984209246	2.500684864
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H	2.072633588	2.523472760	0.109740991
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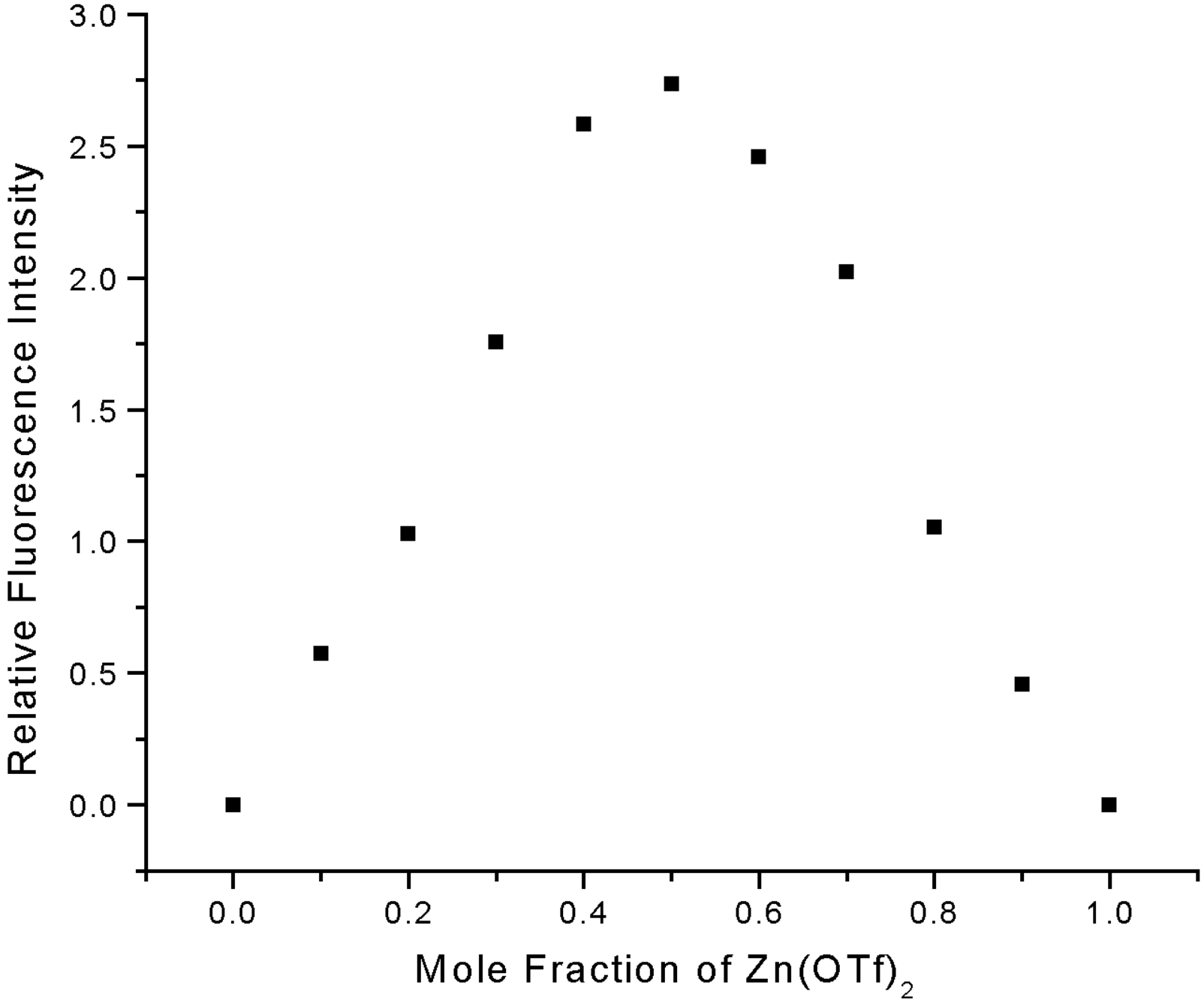
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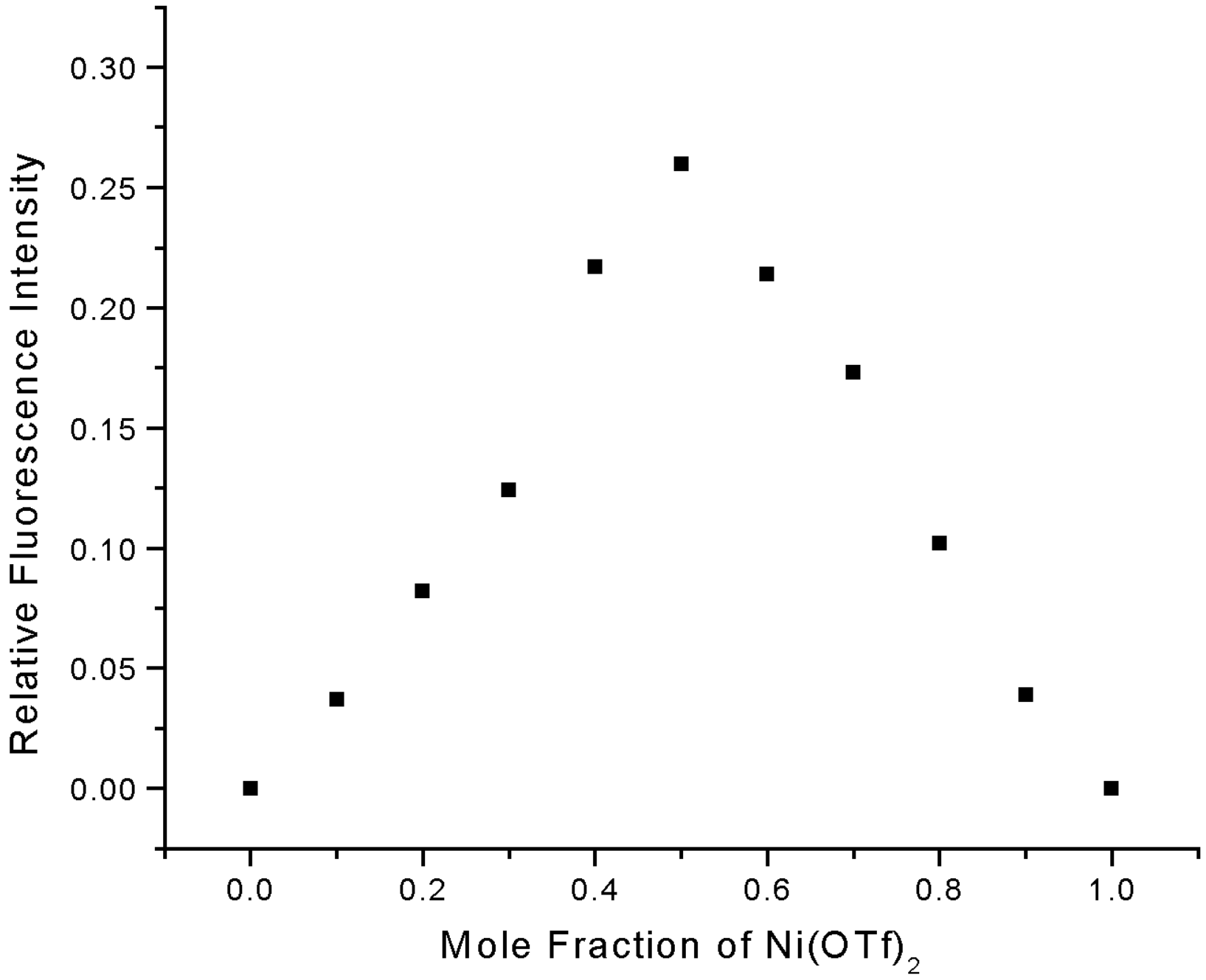
Mole Fraction of Zn(OTf)2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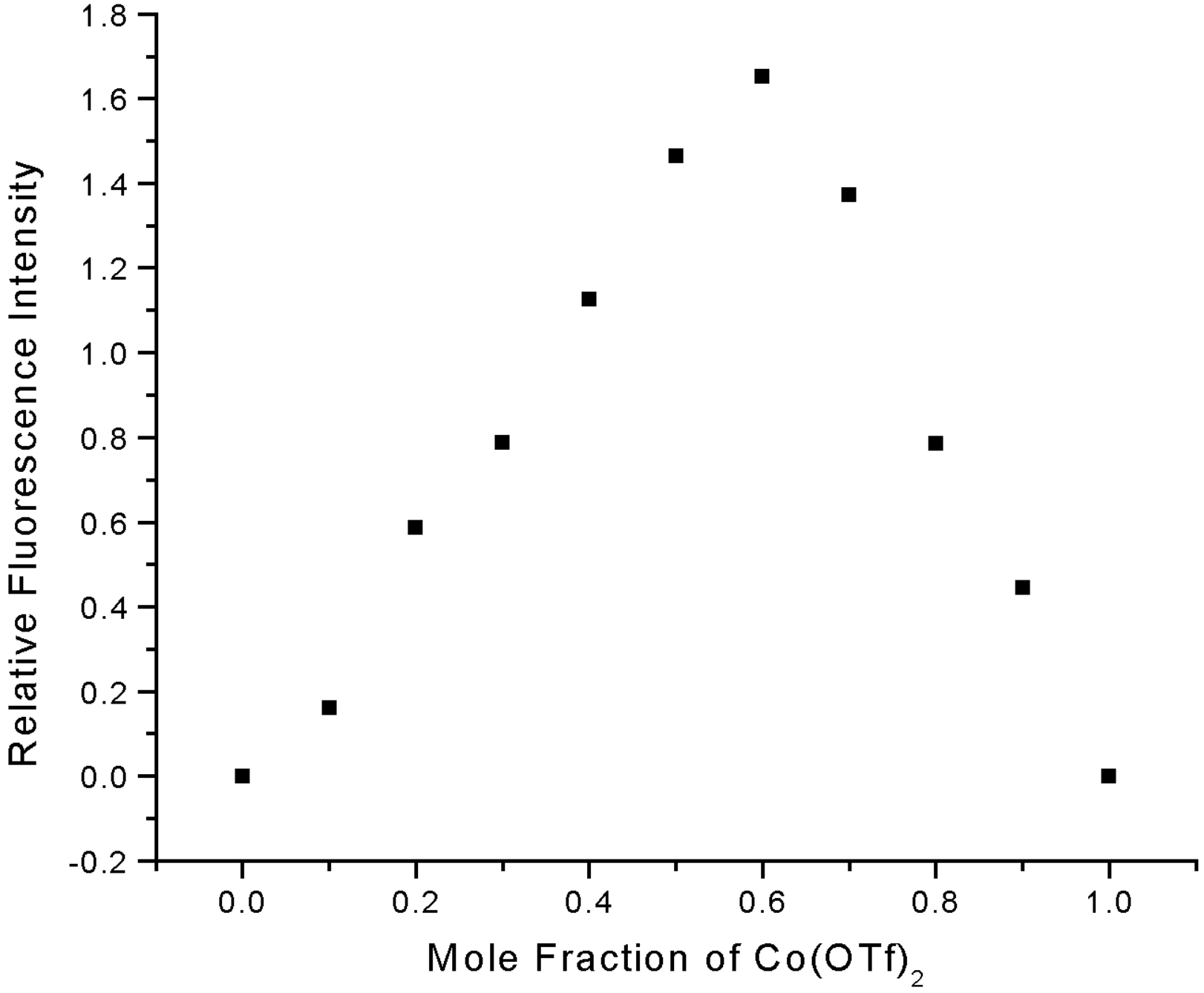
Mole Fraction of Ni(OTf)2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1.0
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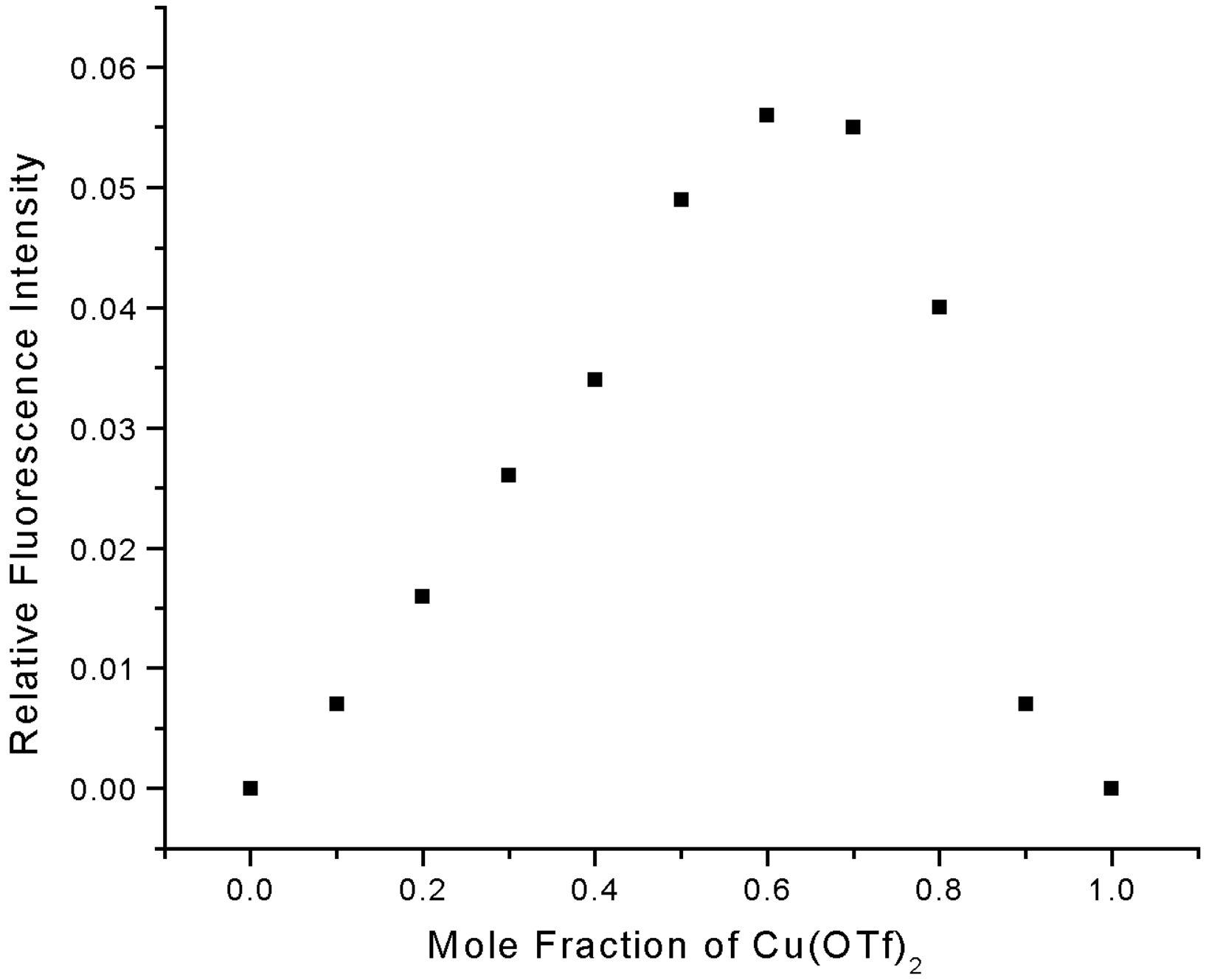
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1.0
Mole Fraction of Cu(OTf)2