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# Visualization of $Zn^{2+}$ Ions in Live Zebrafish Using a Luminescent Iridium(III) Chemosensor

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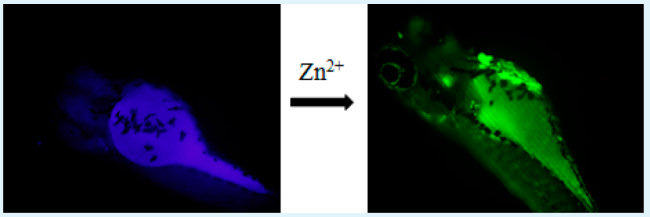
Supporting Information

**ABSTRACT:** A novel luminescent cyclometalated iridium(III) complex-based chemosensor (**1**) bearing a zinc-specific receptor, tris(2-pyridylmethyl)amine, and the 3-phenyl-1*H*-pyrazole ligand has been designed and synthesized. Upon the addition of $Zn^{2+}$ ions to a solution of iridium(III) complex **1**, a pronounced luminescence color change from blue to green can be observed, which may be attributed to the suppression of photoinduced electron transfer upon complexation of complex **1** with $Zn^{2+}$ ions. The interaction of iridium(III) complex **1** with $Zn^{2+}$ ions was investigated by UV−vis absorption titration, emission titration, and $^1$H NMR titration. Furthermore, the iridium(III) complex **1** exhibited good selectivity for $Zn^{2+}$ over 13 other common metal ions, including $K^+$, $Ag^+$, $Na^+$, $Ni^{2+}$, $Fe^{3+}$, $Hg^{2+}$, $Cd^{2+}$, $Mg^{2+}$, $Ca^{2+}$, $Cu^{2+}$, $Mn^{2+}$, $Co^{2+}$, and $Pb^{2+}$ ions. The practical application of the iridium(III) complex **1** in visualizing intracellular $Zn^{2+}$ distribution in live zebrafish was also demonstrated.

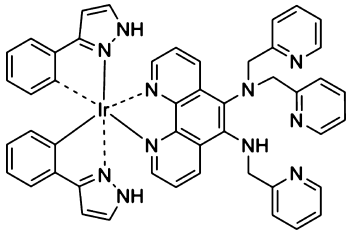



**KEYWORDS:** *iridium(III) complex, zinc ion, chemosensor, luminescent*

## 1. INTRODUCTION

Zinc ions are involved in important physiological functions in living organisms, such as neurotransmission, gene transcription, and immune function.[1] Zinc deficiency is associated with a number of human diseases, including major depressive disorder, developmental defects, chronic liver disease, chronic renal disease, sickle cell disease, and diabetes.[2−4] On the other hand, the elevated consumption of zinc has been implicated in ataxia, lethargy, and copper deficiency.[5,6] Therefore, the accurate detection and imaging of zinc ions is in high demand for the investigation of the regulation of zinc levels and the occurrence of disease states.

A range of detection methods for zinc ion have been developed, including atomic absorption spectrometry[7] and inductively coupled plasma mass spectrometry.[8] However, these methods may be time-consuming and/or require the use of sophisticated instrumentation. The notable advantages of luminescence detection have stimulated the development of a variety of fluorescent chemosensors for zinc ions.[9,10] For example, probes based on zinc-specific receptors conjugated to fluorescent chromophores such as cyanine,[11] fluorescein,[12−23] coumarin,[24−27] 4-nitrobenzoxadiazole,[28−30] quinolone,[31−35] benzoresorufin,[36] 1,8-naphthyridine,[37] methylazacalix[*a*]pyridine,[38] tetraphenylethylene,[39] and rhodamine[40−42] have been reported for $Zn^{2+}$ detection. Some of these fluorescent probes have been used to monitor $Zn^{2+}$ in mitochondria of living cells or in the hippocampus slide.[43,44] However, the use of organic fluorophores in sensing may be limited by their small Stokes shift values and fluorescence lifetimes, as well as autofluorescence arising in biological samples.



**1**

**Figure 1.** Chemical structure of cyclometalated iridium(III) complex **1**.

In this context, transition-metal complexes with metal-to-ligand charge transfer (MLCT) excited states have arisen as viable alternatives to organic fluorophores and have been widely employed for optoelectronic devices, cellular imaging, and

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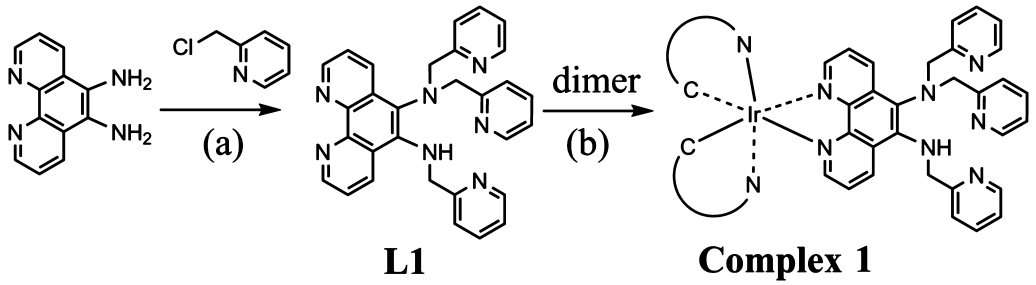
**Scheme 1. Synthetic Pathway of Iridium(III) Complex 1**[a]




[a]Reagents and conditions: (a) picolyl chloride, $CH_3CN$, $K_2CO_3$, $N_2$, reflux, 56.5%; (b) DCM/methanol, reflux.

chemosensing and as structural probes for biomolecules.[45−84] The long phosphorescence lifetime of luminescent metal complexes in the visible region offers a means to eliminate background fluorescence by the use of time-resolved luminescence spectroscopy. A few luminescent iridium(III) complexes have been developed for $Zn^{2+}$ detection, such as the cyclometalated complexes containing the zinc-specific di-2-picolylamine (DPA) receptor as reported by the groups of Lippard, Nam, You, and Lo.[85−88] We herein report the application of a novel luminescent cyclometalated iridium(III) complex containing the $Zn^{2+}$ receptor tris(2-pyridylmethyl)amine (TPA) and the 3-phenyl-1*H*-pyrazole (C^N) ligand for $Zn^{2+}$ ion imaging (Figure 1). Furthermore, the iridium(III) complex **1** was demonstrated to function as a selective luminescent probe for visualizing intracellular $Zn^{2+}$ distribution in live zebrafish.

## 2. EXPERIMENTAL SECTION

**2.1. Materials.** Reagents, unless specified, were purchased from Sigma-Aldrich (St. Louis, MO) and used as received. Iridium chloride hydrate ($IrCl_3{\cdot}xH_2O$) was purchased from Precious Metals Online (Australia).

**2.2. Synthesis of Complex 1.** The precursor iridium(III) dimer complex $Ir_2(C^N)_4Cl_2$ (where C^N = 3-phenyl-1*H*-pyrazole) was prepared according to modified literature methods.[89] A suspension of $Ir_2(C^N)_4Cl_2$ (0.2 mmol) and N^N ligand **L1** (0.44 mmol) in a mixture of DCM/methanol (1:1, 20 mL) was refluxed overnight under a nitrogen atmosphere. The resulting solution was then allowed to cool to room temperature and filtered to remove unreacted cyclometalated dimer. To the filtrate was added an aqueous solution of ammonium hexafluorophosphate (excess), and the filtrate was reduced in volume by rotary evaporation until precipitation of the crude product occurred. The precipiate was then filtered and washed with several portions of water (2 × 50 mL) followed by diethyl ether (2 × 50 mL). The product was recrystallized by acetonitrile/diethyl ether vapor diffusion to yield the title compound.

Data for **L1**: yield 35%; $^1$H NMR (400 MHz, DMSO) $\delta$ 9.00 (dd, $J$ = 4.2, 1.6 Hz, 2H), 8.76−8.56 (m, 4H), 8.50 (ddd, $J$ = 4.8, 1.7, 0.9 Hz,4H), 7.99 (dd, $J$ = 8.2, 1.6 Hz, 4H), 7.80−7.72 (m, 2H), 7.76−7.58 (m, 1H), 7.56−7.37 (m, 2H), 7.30 (ddd, J = 7.6, 4.8, 1.1 Hz, 6H), 6.81 (s, 2H), 4.72 (s, 1H); $^{13}$C NMR (100 MHz, DMSO) $\delta$ 158.93, 149.47, 149.03, 148.42, 145.12, 141.35, 136.77, 136.52, 133.21, 130.33, 130.12, 123.29, 122.32, 122.15, 121.85, 120.94, 120.08, 57.90, 54.96; MALDI-TOF-HRMS $m/z$ calcd 483.2171, found 484.2260.

Data for complex **1**: yield 35%; $^1$H NMR (400 MHz, DMSO) $\delta$ 9.15 (dd, $J$ = 4.2, 1.4 Hz, 4H), 9.07 (dd, $J$ = 4.3, 1.6 Hz, 4H), 8.73−8.66 (m, 4H), 8.63−8.56 (m, 16H), 8.52 (dd, $J$ = 8.1, 1.4 Hz, 4H), 8.30 (d, $J$ = 5.4 Hz, 3H), 8.26 (s, 3H), 8.16 (s, 1H), 8.06 (d, $J$ = 5.4 Hz, 3H), 8.00 (t, $J$ = 6.1 Hz, 8H), 7.88−7.84 (m, 3H), 7.80 (dddd, $J$ = 8.1, 6.2, 4.0, 2.2 Hz, 12H), 7.72−7.60 (m, 17H), 7.40 (ddd, $J$ = 7.6, 4.3, 1.4 Hz, 17H), 7.27 (t, $J$ = 7.6 Hz, 3H), 7.03 (td, $J$ = 6.2, 2.6 Hz, 3H), 6.95 (t, $J$ = 7.6 Hz, 3H), 4.41 (s, 3H), 3.46 (s, 4H); $^{13}$C NMR (100 MHz, DMSO) $\delta$ 164.49, 159.52, 153.31, 151.00, 150.96, 149.10, 147.59, 147.29, 143.53, 136.00, 133.74, 129.17, 127.94, 124.10, 123.38, 123.14, 122.74, 121.49, 119.66, 118.64, 117.43, 108.13, 104.95, 98.06, 44.14, 42.52, 36.48, 30.97; MALDI-TOF-HRMS $m/z$ calcd 966.3332, found 966.1658.

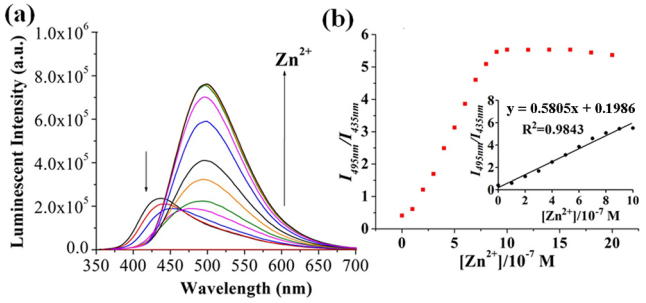



**Figure 2.** (a) Luminescence spectra of **1** (1 $\mu$M) upon addition of various concentrations of $Zn^{2+}$ ions (0−2 $\mu$M) in Tris-buffered solution (25 mM, pH 7.04). (b) Relationship between the luminescence intensity and $Zn^{2+}$ concentration. Inset: luminescence response of **1** at $I_{495nm}/I_{435nm}$ vs $[Zn^{2+}]$. $\lambda_{ex}$ = 320 nm.

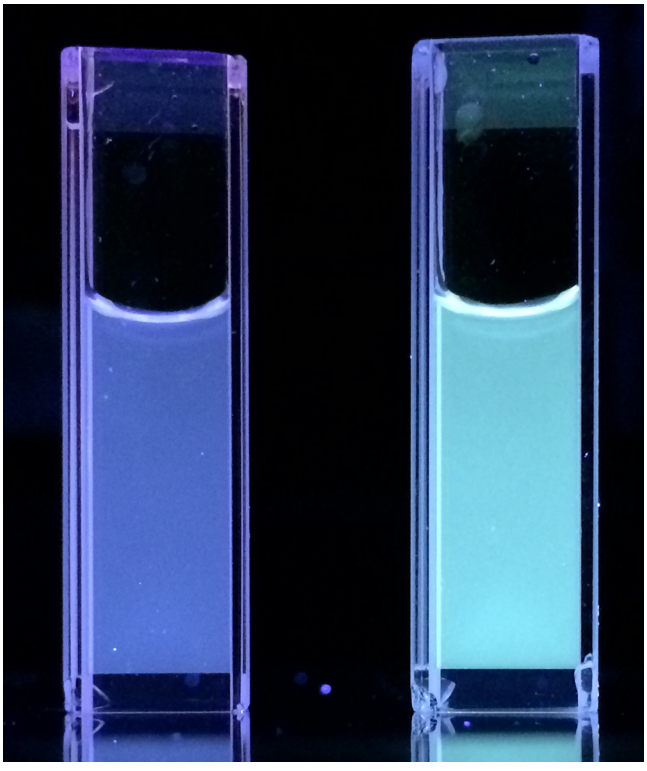


**Figure 3.** Photograph image of complex **1** (1 $\mu$M) in Tris buffer (25 mM, pH 7.04) in the (left) absence or (right) presence of 1 $\mu$M $Zn^{2+}$ under UV illumination.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**2.3. Zinc Ion Imaging in Zebrafish.** Three day old zebrafish were fed with a solution containing 200 $\mu$M $Zn^{2+}$ solution or with a control solution at 28.5 °C for 1.5 h. The zebrafish were washed with PBS solution three times and were incubated with a solution of 40 $\mu$M iridium(III) complex **1** at 28.5 °C for 1 h. Alternatively, $Zn^{2+}$-fed zebrafish were rinsed three times with PBS and then incubated with 50 $\mu$M *N,N,N′,N′*-tetrakis(2-pyridylmethyl)ethylenediamine (TPEN) solution, followed by further rinsing three times with PBS and incubation with 40 $\mu$M iridium(III) complex **1** at 28.5 °C for 1 h. Confocal imaging of zebrafish was performed using a research inverted microscope system (Olympus IX73).

## 3. RESULTS AND DISCUSSION

**3.1. Design and Synthesis of the Iridium(III) Complex.** The iridium(III) complex **1** was designed to bear a N^N ligand (**L1**) incorporating the $Zn^{2+}$ receptor TPA,[90] in addition to two C^N ligands. The coordination of **L1** to $Zn^{2+}$ should influence the MLCT state of the complexes, resulting in observable absorption and emission spectral changes of the complex in the presence of $Zn^{2+}$. Furthermore, the luminescence of the iridium(III) complex is known to be highly sensitive to the nature of the ligands. The iridium(III) complex **1** was prepared according to modified literature procedures (Scheme 1).[89,90]

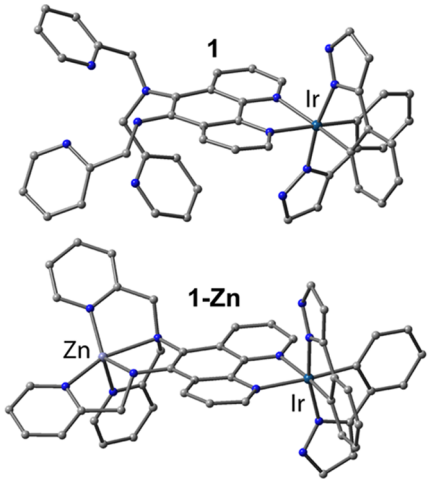


**Figure 4.** DFT-optimized ground-state structures for **1** and **1**−Zn.

**3.2. Photophysical Response of Complex 1 to $Zn^{2+}$.** Complex **1** was titrated with different concentrations of $Zn^{2+}$ in Tris buffer (25 mM, pH 7.04). Interestingly, the luminescence of **1** was significantly enhanced in the presence of increasing concentrations of $Zn^{2+}$ ions, which was accompanied by a substantial red shift ($\Delta\lambda$ = *ca.* 60 nm) in the emission maxima of **1** (Figure 2a). This luminescence enhancement is presumably due to the suppression of photoinduced electron transfer upon complexation of complex **1** with $Zn^{2+}$. A ratiometric ($I_{495nm}/I_{435nm}$) luminescence enhancement of *ca.* 6-fold was observed at 2 $\mu$M $Zn^{2+}$ ions. Furthermore, a linear relationship was observed between the ratiometric enhancement of complex **1** and the $Zn^{2+}$ concentration ($R^2$ = 0.984) in the range of 0−1 $\mu$M $Zn^{2+}$ (Figure 2b). The detection limit at a signal-to-noise ratio of 3 was found to be 36 nM. Furthermore, the presence of $Zn^{2+}$ could be readily observed by the naked eye under UV illumination (Figure 3). In the UV−vis spectrum, the addition of $Zn^{2+}$ to complex **1** led to an increase in absorption at 261 and 298 nm and a decrease in absorption at 281 nm (Figure S1, Supporting Information). Additionally, Job's plot analysis of the luminescence data revealed a maximum in emission enhancement at a 0.5 mole fraction of **1**, indicating a 1:1 stoichiometry between $Zn^{2+}$ ions and **1** (Figure S2, Supporting Information).

**3.3. Theoretical Studies.** Ir(III) complexes are known to possess rich electronic excited states. Time-dependent density functional theory (TD-DFT) calculations have been performed on complex **1** and a hypothetical $Zn^{2+}$-coordinated complex **1**−Zn to investigate the nature of the excited states. The optimized ground-state structures for **1** and **1**−Zn are depicted in Figure 4; their simulation spectra are depicted in Figure S3 (Supporting Information). Because the nature of the transitions in the spectral region of 300−600 nm are quite complex for both complexes, only the major transitions with oscillator strength >0.04 are presented in Figure S3. For complex **1**, the calculated lowest energy absorption bands at around 425 nm are attributed to a mixing of n(amine) → $\pi^*$(phen) and $d_\pi$(Ir) → $\pi^*$(phen) charge transfer transitions (transitions I and II in Figure S4, Supporting Information).

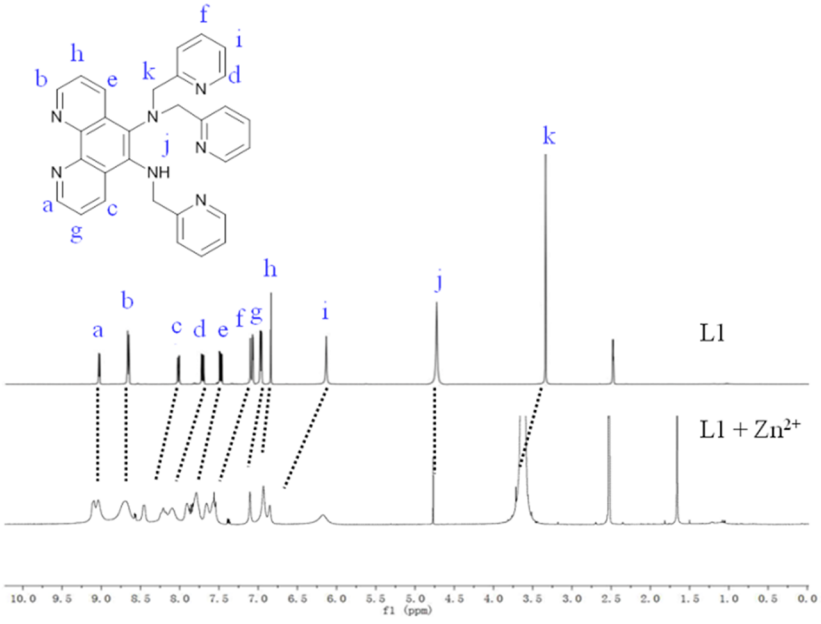


**Figure 5.** $^1$H NMR spectra of **L1** (5 mM) in the (upper) absence and (lower) presence of $ZnCl_2$ (5 mM) in DMSO-$d_6$ at 298 K.

For hypothetical complex **1**−Zn, the calculated lowest energy absorption band at around 475 nm is attributed to the $d_\pi(Ir) \rightarrow \pi^*(phen)$ charge transfer transition (transition I in Figure S5, Supporting Information). The calculated red shift upon $Zn^{2+}$ coordination to **1** is consistent with the experimental finding that a red shift in emission energy is induced upon $Zn^{2+}$ binding to **1**.

**3.4. $^1$H NMR Titration.** To investigate the complexation mode of $Zn^{2+}$ ions, $^1$H NMR titration of **L1** with $Zn^{2+}$ in DMSO-$d_6$ was performed. The results showed several significant spectral changes in the $^1$H NMR spectrum of **L1** upon the addition of $Zn^{2+}$ ions (c, from 7.98 to 8.41 ppm; d, from 7.72 to 8.19 ppm; e, from 7.46 to 7.82 ppm; f, from 7.13 to 7.64 ppm; g, from 6.94 to 7.17 ppm; h, from 6.63 to 6.90 ppm; i, from 6.17 to 6.65 ppm; k, from 3.31 to 3.72 ppm) (Figure 5). The spectral changes suggest that $Zn^{2+}$ ions putatively bind to **L1** via coordination to the nitrogen atoms of the pyridine groups.

**3.5. Selectivity of Iridium(III) Complex 1 for $Zn^{2+}$.** We next evaluated the selectivity of the iridium(III) complex **1** for $Zn^{2+}$ by investigating its luminescence response to 13 other common metal ions, including $Na^+$, $K^+$, $Ag^+$, $Ni^{2+}$, $Fe^{3+}$, $Hg^{2+}$, $Cd^{2+}$, $Mg^{2+}$, $Ca^{2+}$, $Cu^{2+}$, $Mn^{2+}$, $Co^{2+}$, and $Pb^{2+}$ ions. The results showed that only $Zn^{2+}$ ions could significantly enhance the ratiometric luminescence output of complex **1** (Figure 6). By comparison, only minor changes in $I_{495nm}/I_{435nm}$ were observed upon the addition of a 20-fold excess of the $Ag^+$, $Ni^{2+}$, $Fe^{3+}$, $Hg^{2+}$, $Cd^{2+}$, $Mg^{2+}$, $Ca^{2+}$, $Cu^{2+}$, $Mn^{2+}$, $Co^{2+}$, and $Pb^{2+}$ ions. Moreover, complex **1** displayed significant selectivity for $Zn^{2+}$ ions over a 1000-fold excess of $Na^+$ and $K^+$ ions. These results indicate that the iridium(III) complex **1** displays significant selectivity for $Zn^{2+}$ ions over the other metal ions, which originates presumably from the specific interaction of the TPA moiety of complex **1** with $Zn^{2+}$ ions. Additionally, we have investigated the impact of the coaddition of $Cu^{2+}$ ions on the performance of the assay for $Zn^{2+}$ detection. The results showed that addition of 0.2 or 0.4 $\mu$M $Cu^{2+}$ into a 1 $\mu$M $Zn^{2+}$ solution would result in a 24% or 33% decrease in the luminescence of complex **1**, respectively (Figure S6, Supporting Information).

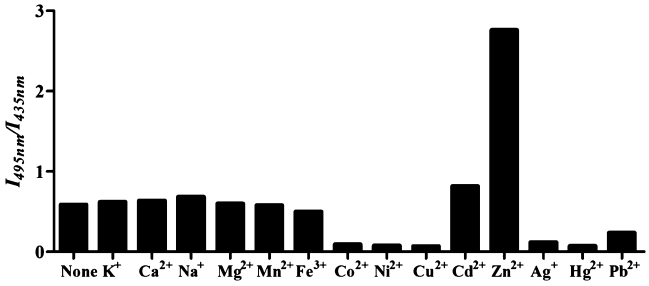


**Figure 6.** Selectivity of complex **1** for $Zn^{2+}$. The concentration of $Zn^{2+}$ ions was 1.0 $\mu$M, the concentrations of $Na^+$ and $K^+$ were 1000 $\mu$M, and the concentrations of the other metal ions were 20.0 $\mu$M.

**3.6. $Zn^{2+}$ Imaging in Zebrafish.** The lifetime of complex **1** was determined to be 4.38 $\mu$s ($\lambda$ = 495 nm), which could make it suitable for $Zn^{2+}$ imaging in biological tissues. Taking advantage of the ratiometric behavior and high sensitivity of complex **1** toward $Zn^{2+}$ ions, the practical application of complex **1** as a luminescent probe to monitor $Zn^{2+}$ distribution in live zebrafish was investigated. Ratiometric imaging revealed a bright green fluorescence in the abdomen of 3 day old zebrafish fed with $Zn^{2+}$, indicating an accumulation of $Zn^{2+}$ ions in the abdomen, while non-$Zn^{2+}$-treated zebrafish appeared blue (Figure 7). Comparison of the luminescence images obtained for zebrafish fed with different concentrations of zinc ions (0, 20 nM, 200 nM, 2 $\mu$M, 20 $\mu$M, and 200 $\mu$M) suggested that down to 200 nM $Zn^{2+}$ ions

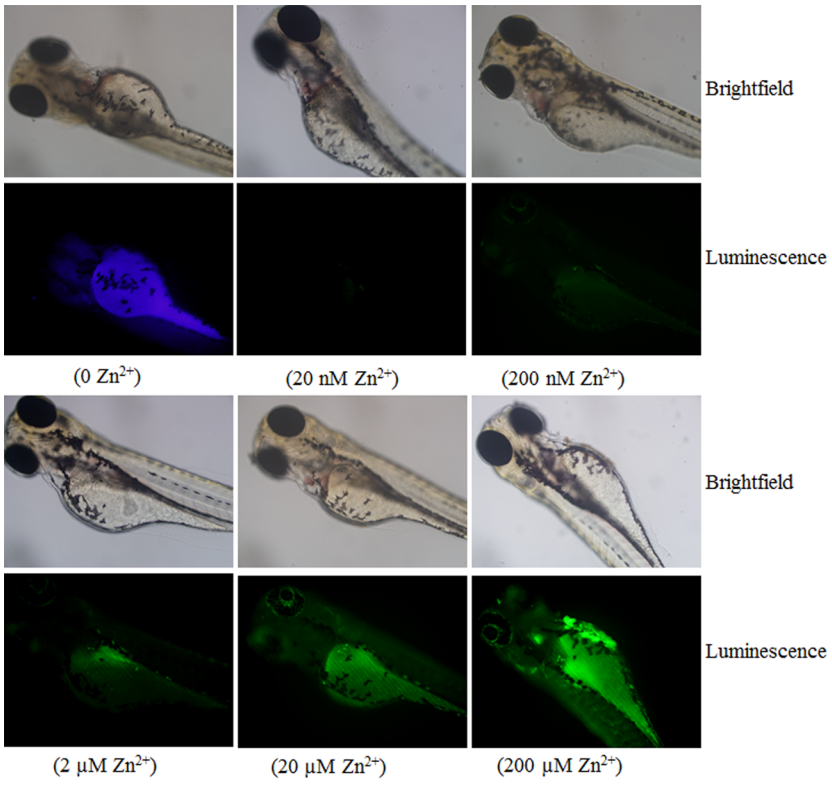


**Figure 7.** $Zn^{2+}$ imaging in 3 day old zebrafish. Bright-field (upper) and luminescence (lower) images of zebrafish fed with different concentrations of $Zn^{2+}$ (0, 20 nM, 200 nM, 2 $\mu$M, 20 $\mu$M, and 200 $\mu$M) for 1.5 h followed by incubation with complex **1** (40 $\mu$M) for 1 h.

could be detected in live zebrafish (Figure 7). A time-course experiment revealed that the green fluorescence intensity of treated zebrafish increased with the $Zn^{2+}$ incubation time (0, 30, 60, and 90 min) (Figure S7, Supporting Information). Additionally, the green fluorescence in $Zn^{2+}$-treated zebrafish was reduced to an almost undetectable level if the chelator TPEN was also present (Figure 8), suggesting that the green emission

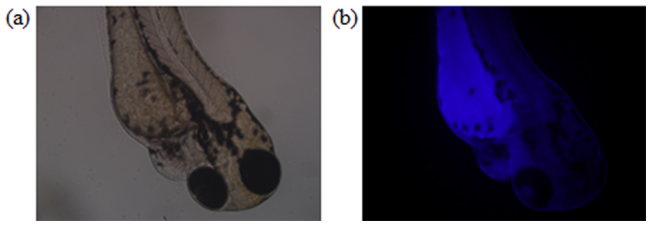


**Figure 8.** (a) Bright-field and (b) luminescence images of a zebrafish incubated with $Zn^{2+}$ (200 $\mu$M) for 1.5 h, followed by treatment with TPEN (50 $\mu$M) for 1.5 h and incubation with complex **1** (40 $\mu$M) for 1 h.

observed in Figure 7 arose from the complexation of complex **1** with $Zn^{2+}$. These results indicate that complex **1** can be used as a luminescent imaging agent for $Zn^{2+}$ ions in live zebrafish.

## 4. CONCLUSION

In summary, we have synthesized a novel luminescent cyclometalated iridium(III) complex containing the zinc-binding TPA group. Complex **1** displayed pronounced luminescence changes upon the addition of $Zn^{2+}$ in aqueous solution and showed good sensitivity and selectivity for $Zn^{2+}$ over 13 other metal ions, including $K^+$, $Ag^+$, $Na^+$, $Ni^{2+}$, $Fe^{3+}$, $Hg^{2+}$, $Cd^{2+}$, $Mg^{2+}$, $Ca^{2+}$, $Cu^{2+}$, $Mn^{2+}$, $Co^{2+}$, and $Pb^{2+}$ ions. Compared to other organic zinc ion chemosensors, which typically require an organic solvent for detection, our developed iridium(III) complex-based $Zn^{2+}$ chemosensor could be used in aqueous buffer and exhibits a higher selectivity against $Cd^{2+}$ ions than that of some other reported $Zn^{2+}$ ion chemosensors. A comparison of recently reported chemosensors for $Zn^{2+}$ ion is presented in Table 1. Furthermore, the practical application of complex **1** for imaging $Zn^{2+}$ ions in live zebrafish was successfully demonstrated. We envision that the iridium(III) complex could be further developed as a useful probe for monitoring zinc ion distribution in biological samples.

**Table 1. Comparison of Chemosensors for $Zn^{2+}$ Ion Assays Recently Reported in the Literature**

| signal output | detection condition | selectivity against $Cd^{2+}$ | real sample application | ref |
|---|---|---|---|---|
| ratiometric and colorimetric | ACN/$H_2O$ (80:20) | good | not reported | 90 |
| fluorescent | HEPES buffer | not good | HeLa cells | 91 |
| fluorescent | ACN/$H_2O$ (18:7) | good | not reported | 92 |
| fluorescent | ACN | good | not reported | 93 |
| fluorescent | DMF/$H_2O$ (1:1) | not good | not reported | 94 |
| fluorescent | EtOH/$H_2O$ (9:1) | not good | HepG-2 cells | 95 |
| fluorescent | EtOH | good | MCF-7 cells | 96 |
| luminescent and ratiometric | Tris buffer | good | zebrafish | this study |

## ■ ASSOCIATED CONTENT

### Ⓢ Supporting Information

General experimental procedures, photophysical measurements, DFT calculations, UV–vis absorption spectra, and Job's plot analysis. This material is available free of charge via the Internet at http://pubs.acs.org.

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### Notes

The authors declare no competing financial interest.

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## ■ REFERENCES

(1) Frederickson, C. J.; Koh, J.-Y.; Bush, A. I. The Neurobiology of Zinc in Health and Disease. *Nat. Rev. Neurosci.* **2005**, *6*, 449–462.
(2) Prasad, A. S. Zinc Deficiency. *BMJ [Br. Med. J.]* **2003**, *326*, 409–410.
(3) Hambidge, M. Human Zinc Deficiency. *J. Nutr.* **2000**, *130*, 1344S–1349S.
(4) Prasad, A. S. Clinical Manifestations of Zinc Deficiency. *Annu. Rev. Nutr.* **1985**, *5*, 341–363.
(5) Sandström, B. Diagnosis of Zinc Deficiency and Excess in Individuals and Populations. *Food Nutr. Bull.* **2001**, *22*, 133–137.
(6) Nriagu, J., Zinc Toxicity in Humans. In *Encyclopedia of Environmental Health*; Nriagu, J. O., Ed.; Elsevier: Burlington, MA, 2011; pp 801–807.
(7) Chen, J.; Teo, K. C. Determination of Cadmium, Copper, Lead and Zinc in Water Samples by Flame Atomic Absorption Spectrometry after Cloud Point Extraction. *Anal. Chim. Acta* **2001**, *450*, 215–222.
(8) Binet, M. R. B.; Ma, R.; McLeod, C. W.; Poole, R. K. Detection and Characterization of Zinc- and Cadmium-Binding Proteins in *Escherichia coli* by Gel Electrophoresis and Laser Ablation-Inductively Coupled Plasma-Mass Spectrometry. *Anal. Biochem.* **2003**, *318*, 30–38.
(9) Li, X.; Gao, X.; Shi, W.; Ma, H. Design Strategies for Water-Soluble Small Molecular Chromogenic and Fluorogenic Probes. *Chem. Rev.* **2013**, *114*, 590–659.
(10) Xu, Z.; Yoon, J.; Spring, D. R. Fluorescent Chemosensors for $Zn^{2+}$. *Chem. Soc. Rev.* **2010**, *39*, 1996–2006.
(11) Guo, Z.; Kim, G.-H.; Shin, I.; Yoon, J. A Cyanine-Based Fluorescent Sensor for Detecting Endogenous Zinc Ions in Live Cells and Organisms. *Biomaterials* **2012**, *33*, 7818–7827.
(12) Chang, C. J.; Nolan, E. M.; Jaworski, J.; Okamoto, K.-I.; Hayashi, Y.; Sheng, M.; Lippard, S. J. ZP8, a Neuronal Zinc Sensor with Improved Dynamic Range; Imaging Zinc in Hippocampal Slices with Two-Photon Microscopy. *Inorg. Chem.* **2004**, *43*, 6774–6779.
(13) Nolan, E. M.; Burdette, S. C.; Harvey, J. H.; Hilderbrand, S. A.; Lippard, S. J. Synthesis and Characterization of Zinc Sensors Based on a Monosubstituted Fluorescein Platform. *Inorg. Chem.* **2004**, *43*, 2624–2635.

(14) Chang, C. J.; Nolan, E. M.; Jaworski, J.; Burdette, S. C.; Sheng, M.; Lippard, S. J. Bright Fluorescent Chemosensor Platforms for Imaging Endogenous Pools of Neuronal Zinc. *Chem. Biol.* **2004**, *11*, 203−210.
(15) Burdette, S. C.; Frederickson, C. J.; Bu, W.; Lippard, S. J. ZP4, an Improved Neuronal $Zn^{2+}$ Sensor of the Zinpyr Family. *J. Am. Chem. Soc.* **2003**, *125*, 1778−1787.
(16) Goldsmith, C. R.; Lippard, S. J. 6-Methylpyridyl for Pyridyl Substitution Tunes the Properties of Fluorescent Zinc Sensors of the Zinpyr Family. *Inorg. Chem.* **2005**, *45*, 555−561.
(17) Nolan, E. M.; Jaworski, J.; Racine, M. E.; Sheng, M.; Lippard, S. J. Midrange Affinity Fluorescent Zn(II) Sensors of the Zinpyr Family: Syntheses, Characterization, and Biological Imaging Applications. *Inorg. Chem.* **2006**, *45*, 9748−9757.
(18) Hirano, T.; Kikuchi, K.; Urano, Y.; Higuchi, T.; Nagano, T. Highly Zinc-Selective Fluorescent Sensor Molecules Suitable for Biological Applications. *J. Am. Chem. Soc.* **2000**, *122*, 12399−12400.
(19) Burdette, S. C.; Lippard, S. J. The Rhodafluor Family. An Initial Study of Potential Ratiometric Fluorescent Sensors for $Zn^{2+}$. *Inorg. Chem.* **2002**, *41*, 6816−6823.
(20) Burdette, S. C.; Walkup, G. K.; Spingler, B.; Tsien, R. Y.; Lippard, S. J. Fluorescent Sensors for $Zn^{2+}$ Based on a Fluorescein Platform: Synthesis, Properties and Intracellular Distribution. *J. Am. Chem. Soc.* **2001**, *123*, 7831−7841.
(21) Walkup, G. K.; Burdette, S. C.; Lippard, S. J.; Tsien, R. Y. A New Cell-Permeable Fluorescent Probe for $Zn^{2+}$. *J. Am. Chem. Soc.* **2000**, *122*, 5644−5645.
(22) Wong, B. A.; Friedle, S.; Lippard, S. J. Subtle Modification of 2,2-Dipicolylamine Lowers the Affinity and Improves the Turn-On of Zn(II)-Selective Fluorescent Sensors. *Inorg. Chem.* **2009**, *48*, 7009−7011.
(23) Zhang, X.-A.; Hayes, D.; Smith, S. J.; Friedle, S.; Lippard, S. J. New Strategy for Quantifying Biological Zinc by a Modified Zinpyr Fluorescence Sensor. *J. Am. Chem. Soc.* **2008**, *130*, 15788−15789.
(24) Komatsu, K.; Urano, Y.; Kojima, H.; Nagano, T. Development of an Iminocoumarin-Based Zinc Sensor Suitable for Ratiometric Fluorescence Imaging of Neuronal Zinc. *J. Am. Chem. Soc.* **2007**, *129*, 13447−13454.
(25) Mizukami, S.; Okada, S.; Kimura, S.; Kikuchi, K. Design and Synthesis of Coumarin-Based $Zn^{2+}$ Probes for Ratiometric Fluorescence Imaging. *Inorg. Chem.* **2009**, *48*, 7630−7638.
(26) Xu, Z.; Liu, X.; Pan, J.; Spring, D. R. Coumarin-Derived Transformable Fluorescent Sensor for $Zn^{2+}$. *Chem. Commun.* **2012**, *48*, 4764−4766.
(27) An, J.-M.; Yan, M.-H.; Yang, Z.-Y.; Li, T.-R.; Zhou, Q.-X. A Turn-On Fluorescent Sensor for Zn(II) Based on Fluorescein-Coumarin Conjugate. *Dyes Pigm.* **2013**, *99*, 1−5.
(28) Jiang, W.; Fu, Q.; Fan, H.; Wang, W. An NBD Fluorophore-Based Sensitive and Selective Fluorescent Probe for Zinc Ion. *Chem. Commun.* **2008**, 259−261.
(29) Qian, F.; Zhang, C.; Zhang, Y.; He, W.; Gao, X.; Hu, P.; Guo, Z. Visible Light Excitable $Zn^{2+}$ Fluorescent Sensor Derived from an Intramolecular Charge Transfer Fluorophore and Its in Vitro and in Vivo Application. *J. Am. Chem. Soc.* **2009**, *131*, 1460−1468.
(30) Xu, Z.; Kim, G.-H.; Han, S. J.; Jou, M. J.; Lee, C.; Shin, I.; Yoon, J. An NBD-Based Colorimetric and Fluorescent Chemosensor for $Zn^{2+}$ and Its Use for Detection of Intracellular Zinc Ions. *Tetrahedron* **2009**, *65*, 2307−2312.
(31) Dhara, K.; Karan, S.; Ratha, J.; Roy, P.; Chandra, G.; Manassero, M.; Mallik, B.; Banerjee, P. A Two-Dimensional Coordination Compound as a Zinc Ion Selective Luminescent Probe for Biological Applications. *Chem.—Asian J.* **2007**, *2*, 1091−1100.
(32) Liu, Y.; Zhang, N.; Chen, Y.; Wang, L.-H. Fluorescence Sensing and Binding Behavior of Aminobenzenesulfonamidoquinolino-$\beta$-cyclodextrin to $Zn^{2+}$. *Org. Lett.* **2006**, *9*, 315−318.
(33) Zhang, Y.; Guo, X.; Si, W.; Jia, L.; Qian, X. Ratiometric and Water-Soluble Fluorescent Zinc Sensor of Carboxamidoquinoline with an Alkoxyethylamino Chain as Receptor. *Org. Lett.* **2008**, *10*, 473−476.
(34) Liu, Z.; Zhang, C.; Li, Y.; Wu, Z.; Qian, F.; Yang, X.; He, W.; Gao, X.; Guo, Z. A $Zn^{2+}$ Fluorescent Sensor Derived from 2-(Pyridin-2-yl)benzoimidazole with Ratiometric Sensing Potential. *Org. Lett.* **2009**, *11*, 795−798.
(35) Mikata, Y.; Yamashita, A.; Kawamura, A.; Konno, H.; Miyamoto, Y.; Tamotsu, S. Bisquinoline-Based Fluorescent Zinc Sensors. *Dalton Trans.* **2009**, 3800−3806.
(36) Lin, W.; Buccella, D.; Lippard, S. J. Visualization of Peroxynitrite-Induced Changes of Labile $Zn^{2+}$ in the Endoplasmic Reticulum with Benzoresorufin-Based Fluorescent Probes. *J. Am. Chem. Soc.* **2013**, *135*, 13512−13520.
(37) Yu, M.-M.; Li, Z.-X.; Wei, L.-H.; Wei, D.-H.; Tang, M.-S. A 1,8-Naphthyridine-Based Fluorescent Chemodosimeter for the Rapid Detection of $Zn^{2+}$ and $Cu^{2+}$. *Org. Lett.* **2008**, *10*, 5115−5118.
(38) Yang, X.-B.; Yang, B.-X.; Ge, J.-F.; Xu, Y.-J.; Xu, Q.-F.; Liang, J.; Lu, J.-M. Benzo[*a*]phenoxazinium-Based Red-Emitting Chemosensor for Zinc Ions in Biological Media. *Org. Lett.* **2011**, *13*, 2710−2713.
(39) Sun, F.; Zhang, G.; Zhang, D.; Xue, L.; Jiang, H. Aqueous Fluorescence Turn-On Sensor for $Zn^{2+}$ with a Tetraphenylethylene Compound. *Org. Lett.* **2011**, *13*, 6378−6381.
(40) Tomat, E.; Lippard, S. J. Ratiometric and Intensity-Based Zinc Sensors Built on Rhodol and Rhodamine Platforms. *Inorg. Chem.* **2010**, *49*, 9113−9115.
(41) Sivaraman, G.; Anand, T.; Chellappa, D. Turn-On Fluorescent Chemosensor for Zn(II) via Ring Opening of Rhodamine Spirolactam and Their Live Cell Imaging. *Analyst* **2012**, *137*, 5881−5884.
(42) Han, Z.-X.; Zhang, X.-B.; Li, Z.; Gong, Y.-J.; Wu, X.-Y.; Jin, Z.; He, C.-M.; Jian, L.-X.; Zhang, J.; Shen, G.-L.; Yu, R.-Q. Efficient Fluorescence Resonance Energy Transfer-Based Ratiometric Fluorescent Cellular Imaging Probe for $Zn^{2+}$ Using a Rhodamine Spirolactam as a Trigger. *Anal. Chem.* **2010**, *82*, 3108−3113.
(43) Masanta, G.; Lim, C. S.; Kim, H. J.; Han, J. H.; Kim, H. M.; Cho, B. R. A Mitochondrial-Targeted Two-Photon Probe for Zinc Ion. *J. Am. Chem. Soc.* **2011**, *133*, 5698−5700.
(44) Komatsu, K.; Kikuchi, K.; Kojima, H.; Urano, Y.; Nagano, T. Selective Zinc Sensor Molecules with Various Affinities for $Zn^{2+}$, Revealing Dynamics and Regional Distribution of Synaptically Released $Zn^{2+}$ in Hippocampal Slices. *J. Am. Chem. Soc.* **2005**, *127*, 10197−10204.
(45) Wong, W.-Y.; Ho, C.-L. Organometallic Photovoltaics: A New and Versatile Approach for Harvesting Solar Energy Using Conjugated Polymetallaynes. *Acc. Chem. Res.* **2010**, *43*, 1246−1256.
(46) Lamansky, S.; Djurovich, P.; Murphy, D.; Abdel-Razzaq, F.; Lee, H.-E.; Adachi, C.; Burrows, P. E.; Forrest, S. R.; Thompson, M. E. Highly Phosphorescent Bis-Cyclometalated Iridium Complexes: Synthesis, Photophysical Characterization, and Use in Organic Light Emitting Diodes. *J. Am. Chem. Soc.* **2001**, *123*, 4304−4312.
(47) Hanson, K.; Tamayo, A.; Diev, V. V.; Whited, M. T.; Djurovich, P. I.; Thompson, M. E. Efficient Dipyrrin-Centered Phosphorescence at Room Temperature from Bis-Cyclometalated Iridium(III) Dipyrrinato Complexes. *Inorg. Chem.* **2010**, *49*, 6077−6084.
(48) Gill, M. R.; Garcia-Lara, J.; Foster, S. J.; Smythe, C.; Battaglia, G.; Thomas, J. A. A Ruthenium(II) Polypyridyl Complex for Direct Imaging of DNA Structure in Living Cells. *Nat. Chem.* **2009**, *1*, 662−667.
(49) Zhao, Q.; Huang, C.; Li, F. Phosphorescent Heavy-Metal Complexes for Bioimaging. *Chem. Soc. Rev.* **2011**, *40*, 2508−2524.
(50) Gill, M. R.; Thomas, J. A. Ruthenium(II) Polypyridyl Complexes and DNA—From Structural Probes to Cellular Imaging and Therapeutics. *Chem. Soc. Rev.* **2012**, *41*, 3179−3192.
(51) Ahmad, H.; Hazel, B. W.; Meijer, A. J. H. M.; Thomas, J. A.; Wilkinson, K. A. A Self-Assembled Luminescent Host That Selectively Senses ATP in Water. *Chem.—Eur. J.* **2013**, *19*, 5081−5087.
(52) Yang, Y.; Zhao, Q.; Feng, W.; Li, F. Luminescent Chemodosimeters for Bioimaging. *Chem. Rev.* **2013**, *113*, 192−270.
(53) Zhao, Q.; Li, F.; Huang, C. Phosphorescent Chemosensors Based on Heavy-Metal Complexes. *Chem. Soc. Rev.* **2010**, *39*, 3007−3030.
(54) Ma, D.-L.; Wong, W.-L.; Chung, W.-H.; Chan, F.-Y.; So, P.-K.; Lai, T.-S.; Zhou, Z.-Y.; Leung, Y.-C.; Wong, K.-Y. A Highly Selective Luminescent Switch-On Probe for Histidine/Histidine-Rich Proteins and Its Application in Protein Staining. *Angew. Chem., Int. Ed.* **2008**, *47*, 3735−3739.

(55) Li, C.; Yu, M.; Sun, Y.; Wu, Y.; Huang, C.; Li, F. A Nonemissive Iridium(III) Complex That Specifically Lights-Up the Nuclei of Living Cells. *J. Am. Chem. Soc.* **2011**, *133*, 11231−11239.
(56) Li, C.; Liu, Y.; Wu, Y.; Sun, Y.; Li, F. The Cellular Uptake and Localization of Non-Emissive Iridium(III) Complexes as Cellular Reaction-Based Luminescence Probes. *Biomaterials* **2013**, *34*, 1223−1234.
(57) Lo, K. K.-W.; Zhang, K. Y.; Leung, S.-K.; Tang, M.-C. Exploitation of the Dual-Emissive Properties of Cyclometalated Iridium(III)−Polypyridine Complexes in the Development of Luminescent Biological Probes. *Angew. Chem., Int. Ed.* **2008**, *47*, 2213−2216.
(58) Guerchais, V.; Fillaut, J.-L. Sensory Luminescent Iridium(III) and Platinum(II) Complexes for Cation Recognition. *Coord. Chem. Rev.* **2011**, *255*, 2448−2457.
(59) Brandel, J.; Sairenji, M.; Ichikawa, K.; Nabeshima, T. Remarkable $Mg^{2+}$-Selective Emission of an Azacrown Receptor Based on Ir(III) Complex. *Chem. Commun.* **2010**, *46*, 3958−3960.
(60) Li, G.; Chen, Y.; Wu, J.; Ji, L.; Chao, H. Thiol-Specific Phosphorescent Imaging in Living Cells with an Azobis(2,2′-bipyridine)-Bridged Dinuclear Iridium(III) Complex. *Chem. Commun.* **2013**, *49*, 2040−2042.
(61) Shu, Q.; Birlenbach, L.; Schmittel, M. A Bis(ferrocenyl)-phenanthroline Iridium(III) Complex as a Lab-on-a-Molecule for Cyanide and Fluoride in Aqueous Solution. *Inorg. Chem.* **2012**, *51*, 13123−13127.
(62) Qinghai, S.; Bats, J. W.; Schmittel, M. Two Closely Related Iridium(III) Complexes as Colorimetric and Fluorometric Chemodosimeters for Nitrite in Aqueous Solution Operating along Different Modes of Action. *Inorg. Chem.* **2011**, *50*, 10531−10533.
(63) Shi, H.-F.; Liu, S.-J.; Sun, H.-B.; Xu, W.-J.; An, Z.-F.; Chen, J.; Sun, S.; Lu, X.-M.; Zhao, Q.; Huang, W. Simple Conjugated Polymers with On-Chain Phosphorescent Iridium(III) Complexes: Toward Ratiometric Chemodosimeters for Detecting Trace Amounts of Mercury(II). *Chem.—Eur. J.* **2010**, *16*, 12158−12167.
(64) Hou, X.-G.; Wu, Y.; Cao, H.-T.; Sun, H.-Z.; Li, H.-B.; Shan, G.-G.; Su, Z.-M. A Cationic Iridium(III) Complex with Aggregation-Induced Emission (AIE) Properties for Highly Selective Detection of Explosives. *Chem. Commun.* **2014**, *50*, 6031−6034.
(65) Chen, K.; Schmittel, M. A Triple-Channel Lab-on-a-Molecule for Triple-Anion Quantification Using an Iridium(III)-Imidazolium Conjugate. *Chem. Commun.* **2014**, *50*, 5756−5759.
(66) Steunenberg, P.; Ruggi, A.; van den Berg, N. S.; Buckle, T.; Kuil, J.; van Leeuwen, F. W. B.; Velders, A. H. Phosphorescence Imaging of Living Cells with Amino Acid-Functionalized Tris(2-phenylpyridine)-iridium(III) Complexes. *Inorg. Chem.* **2012**, *51*, 2105−2114.
(67) Li, S. P.-Y.; Lau, C. T.-S.; Louie, M.-W.; Lam, Y.-W.; Cheng, S. H.; Lo, K. K.-W. Mitochondria-Targeting Cyclometalated Iridium(III)−PEG Complexes with Tunable Photodynamic Activity. *Biomaterials* **2013**, *34*, 7519−7532.
(68) Lo, K. K.-W.; Chan, B. T.-N.; Liu, H.-W.; Zhang, K. Y.; Li, S. P.-Y.; Tang, T. S.-M. Cyclometalated Iridium(III) Polypyridine Dibenzocyclooctyne Complexes as the First Phosphorescent Bioorthogonal Probes. *Chem. Commun.* **2013**, *49*, 4271−4273.
(69) Zhang, K. Y.; Liu, H.-W.; Fong, T. T.-H.; Chen, X.-G.; Lo, K. K.-W. Luminescent Dendritic Cyclometalated Iridium(III) Polypyridine Complexes: Synthesis, Emission Behavior, and Biological Properties. *Inorg. Chem.* **2010**, *49*, 5432−5443.
(70) Leung, S.-K.; Liu, H.-W.; Lo, K. K.-W. Functionalization of Luminescent Cyclometalated Iridium(III) Polypyridine Complexes with a Fluorous Moiety: Photophysics, Protein-Binding, Bioconjugation, and Cellular Uptake Properties. *Chem. Commun.* **2011**, *47*, 10548−10550.
(71) You, Y. Phosphorescence Bioimaging Using Cyclometalated Ir(III) Complexes. *Curr. Opin. Chem. Biol.* **2013**, *17*, 699−707.
(72) Han, Y.; You, Y.; Lee, Y.-M.; Nam, W. Double Action: Toward Phosphorescence Ratiometric Sensing of Chromium Ion. *Adv. Mater.* **2012**, *24*, 2748−2754.
(73) You, Y.; Han, Y.; Lee, Y.-M.; Park, S. Y.; Nam, W.; Lippard, S. J. Phosphorescent Sensor for Robust Quantification of Copper(II) Ion. *J. Am. Chem. Soc.* **2011**, *133*, 11488−11491.
(74) Ma, D.-L.; Xu, T.; Chan, D. S.-H.; Man, B. Y.-W.; Fong, W.-F.; Leung, C.-H. A Highly Selective, Label-free, Homogenous Luminescent Switch-On Probe for the Detection of Nanomolar Transcription Factor NF-kappaB. *Nucleic Acids Res.* **2011**, *39*, e67.
(75) Man, B. Y.-W.; Chan, H.-M.; Leung, C.-H.; Chan, D. S.-H.; Bai, L.-P.; Jiang, Z.-H.; Li, H.-W.; Ma, D.-L. Group 9 Metal-Based Inhibitors of [Small Beta]-Amyloid (1-40) Fibrillation as Potential Therapeutic Agents for Alzheimer's Disease. *Chemical Sci.* **2011**, *2*, 917−921.
(76) He, H.-Z.; Chan, D. S.-H.; Leung, C.-H.; Ma, D.-L. A Highly Selective G-quadruplex-Based Luminescent Switch-On Probe for the Detection of Gene Deletion. *Chem. Commun.* **2012**, *48*, 9462−9464.
(77) Chan, D. S.-H.; Fu, W.-C.; Wang, M.; Liu, L.-J.; Leung, C.-H.; Ma, D.-L. A Highly Selective and Non-Reaction Based Chemosensor for the Detection of $Hg^{2+}$ Ions Using a Luminescent Iridium(III) Complex. *PLoS ONE* **2013**, *8*, e60114.
(78) He, H.-Z.; Leung, K.-H.; Yang, H.; Chan, D. S.-H.; Leung, C.-H.; Zhou, J.; Bourdoncle, A.; Mergny, J.-L.; Ma, D.-L. Label-Free Detection of Sub-Nanomolar Lead(II) Ions in Aqueous Solution Using a Metal-Based Luminescent Switch-On Probe. *Biosens. Bioelectron.* **2013**, *41*, 871−874.
(79) He, H.-Z.; Wang, M.; Chan, D. S.-H.; Leung, C.-H.; Qiu, J.-W.; Ma, D.-L. A Label-Free G-quadruplex-Based Luminescent Switch-On Assay for the Selective Detection of Histidine. *Methods* **2013**, *64*, 205−211.
(80) Leung, K.-H.; He, H.-Z.; Ma, V. P.-Y.; Chan, D. S.-H.; Leung, C.-H.; Ma, D.-L. A Luminescent G-quadruplex Switch-On Probe for the Highly Selective and Tunable Detection of Cysteine and Glutathione. *Chem. Commun.* **2013**, *49*, 771−773.
(81) Leung, K.-H.; He, H.-Z.; Ma, V. P.-Y.; Zhong, H.-J.; Chan, D. S.-H.; Zhou, J.; Mergny, J.-L.; Leung, C.-H.; Ma, D.-L. Detection of Base Excision Repair Enzyme Activity Using a Luminescent G-quadruplex Selective Switch-On Probe. *Chem. Commun.* **2013**, *49*, 5630−5632.
(82) Leung, K.-H.; He, H.-Z.; Wang, W.; Zhong, H.-J.; Chan, D. S.-H.; Leung, C.-H.; Ma, D.-L. Label-Free Luminescent Switch-On Detection of Endonuclease IV Activity Using a G-Quadruplex-Selective Iridium(III) Complex. *ACS. Appl. Mater. Interfaces* **2013**, *5*, 12249−12253.
(83) Leung, K.-H.; Lu, L.; Wang, M.; Mak, T.-Y.; Chan, D. S.-H.; Tang, F.-K.; Leung, C.-H.; Kwan, H.-Y.; Yu, Z.; Ma, D.-L. A Label-Free Luminescent Switch-On Assay for ATP Using a G-Quadruplex-Selective Iridium(III) Complex. *PLoS ONE* **2013**, *8*, e77021.
(84) He, H.-Z.; Leung, K.-H.; Wang, W.; Chan, D. S.-H.; Leung, C.-H.; Ma, D.-L. Label-Free Luminescence Switch-On Detection of T4 Polynucleotide Kinase Activity Using a G-quadruplex-Selective Probe. *Chem. Commun.* **2014**, *50*, 5313−5315.
(85) You, Y.; Lee, S.; Kim, T.; Ohkubo, K.; Chae, W.-S.; Fukuzumi, S.; Jhon, G.-J.; Nam, W.; Lippard, S. J. Phosphorescent Sensor for Biological Mobile Zinc. *J. Am. Chem. Soc.* **2011**, *133*, 18328−18342.
(86) Woo, H.; Cho, S.; Han, Y.; Chae, W.-S.; Ahn, D.-R.; You, Y.; Nam, W. Synthetic Control over Photoinduced Electron Transfer in Phosphorescence Zinc Sensors. *J. Am. Chem. Soc.* **2013**, *135*, 4771−4787.
(87) Lee, P.-K.; Law, W. H.-T.; Liu, H.-W.; Lo, K. K.-W. Luminescent Cyclometalated Iridium(III) Polypyridine Di-2-picolylamine Complexes: Synthesis, Photophysics, Electrochemistry, Cation Binding, Cellular Internalization, and Cytotoxic Activity. *Inorg. Chem.* **2011**, *50*, 8570−8579.
(88) You, Y.; Cho, S.; Nam, W. Cyclometalated Iridium(III) Complexes for Phosphorescence Sensing of Biological Metal Ions. *Inorg. Chem.* **2013**, *53*, 1804−1815.
(89) Lamansky, S.; Djurovich, P.; Murphy, D.; Abdel-Razzaq, F.; Kwong, R.; Tsyba, I.; Bortz, M.; Mui, B.; Bau, R.; Thompson, M. E. Synthesis and Characterization of Phosphorescent Cyclometalated Iridium Complexes. *Inorg. Chem.* **2001**, *40*, 1704−1711.
(90) Xu, Z.; Qian, X.; Cui, J.; Zhang, R. Exploiting the Deprotonation Mechanism for the Design of Ratiometric and Colorimetric $Zn^{2+}$

Fluorescent Chemosensor with a Large Red-Shift in Emission. *Tetrahedron* **2006**, *62*, 10117−10122.
(91) Hanaoka, K.; Muramatsu, Y.; Urano, Y.; Terai, T.; Nagano, T. Design and Synthesis of a Highly Sensitive Off−On Fluorescent Chemosensor for Zinc Ions Utilizing Internal Charge Transfer. *Chem.—Eur. J.* **2010**, *16*, 568−572.
(92) Gong, H.-Y.; Zheng, Q.-Y.; Zhang, X.-H.; Wang, D.-X.; Wang, M.-X. Methylazacalix[4]pyridine: En Route to $Zn^{2+}$-Specific Fluoresence Sensors. *Org. Lett.* **2006**, *8*, 4895−4898.
(93) Wu, J.-S.; Liu, W.-M.; Zhuang, X.-Q.; Wang, F.; Wang, P.-F.; Tao, S.-L.; Zhang, X.-H.; Wu, S.-K.; Lee, S.-T. Fluorescence Turn On of Coumarin Derivatives by Metal Cations: A New Signaling Mechanism Based on C═N Isomerization. *Org. Lett.* **2006**, *9*, 33−36.
(94) Mikata, Y.; Wakamatsu, M.; Yano, S. Tetrakis(2-quinolinylmethyl)ethylenediamine (TQEN) as a New Fluorescent Sensor for Zinc. *Dalton Trans.* **2005**, 545−550.
(95) Zhang, B.; Cao, K.-S.; Xu, Z.-A.; Yang, Z.-Q.; Chen, H.-W.; Huang, W.; Yin, G.; You, X.-Z. Cell-Compatible Fluorescent Chemosensor for $Zn^{2+}$ Based on a 3,8-Extended 1,10-Phenanthroline Derivative. *Eur. J. Inorg. Chem.* **2012**, *2012*, 3844−3851.
(96) Li, Z.; Yu, M.; Zhang, L.; Yu, M.; Liu, J.; Wei, L.; Zhang, H. A "Switching On" Fluorescent Chemodosimeter of Selectivity to $Zn^{2+}$ and Its Application to MCF-7 Cells. *Chem. Commun.* **2010**, *46*, 7169−7171.