Electrogenerated chemiluminescence of cyclometalated iridium (III) complexes with derived β -acetylacetone as ancillary ligand

Shunqin Xia, Danjuan Han, Hongfang Gao, Ying Zhao, Honglan Qi*, Chengxiao Zhang*

Key Laboratory of Analytical Chemistry for Life Science of Shaanxi Province, School of Chemistry and Chemical Engineering, Shaanxi Normal University, Xi'an 710062, PR China

* To whom correspondence should be addressed. E-mail: honglanqi@snnu.edu.cn (HL Qi); cxzhang@snnu.edu.cn (CX Zhang); Phone: +86 29 81530726; Fax: +86-29-81530727.

Abstract

Two cyclometalated iridium(III) complexes, (pq)₂Ir(sa) and (pq)₂Ir(psa), with 2-phenylquinoline (pq) as the C^N main ligand and succinylacetone (sa) or 4,6-dioxo-6-phenylhexanoic acid (psa) as the O^O ancillary ligand, were synthesized at first time. The solubility was reached down to 0.1 mM for (pq)₂Ir(sa) and 10 μM for (pq)₂Ir(psa) in acetonitrile-water (v/v=5:95) solution. The photophysical, electrochemical and electrogenerated chemiluminescence (ECL) properties of (pq)₂Ir(sa) and (pq)₂Ir(psa) in acetonitrile were mainly investigated. The oxidation peak potential of (pq)₂Ir(sa) and (pq)₂Ir(psa) appears at +0.79V and +0.8 V vs SCE. (pq)₂Ir(sa) and (pq)₂Ir(psa) display orange ECL emission band with a maximum wavelength at 621 nm and 610 nm, respectively. ECL efficiencies of (pq)₂Ir(sa) and (pq)₂Ir(psa) in acetonitrile containing tripropylamine were calculated to be 21.3-folds and 35.4-folds higher than that of Ru(bpy)₃²⁺. It was surprised to find that small amount of water can greatly increase the ECL intensity of these two complexes in acetonitrile containing tripropylamine. The ECL methods for the determination of trace water in organic solvent were developed in the range from 0.02 to 4.0% (v/v) and 0.05 to 4.0% (v/v) with detection limit of 0.01% and 0.03% for (pq)₂Ir(sa) and (pq)₂Ir(psa), respectively. It was found that the O^O ancillary ligand played a key function in the water-enhanced ECL emission. A double coreactant ECL pathway is proposed for the water-enhanced ECL emission. This work demonstrates that (pq)₂Ir(sa) and (pq)₂Ir(psa) have lower potential and high ECL coefficient, which are promising for the effective use as ECL emitter in analytical application.

Keywords: Electrogenated chemiluminescence; Iridium(III) complex; electrochemistry; water

Introduction

Electrogenerated chemiluminescence (ECL) was first identified in the 1960s through the use of organic compounds, such as, rubrene, 9,10-diphenylanthracene (DPA), and related compounds [1, 2]. The first ECL of metal complexes tris-(2,2'-bipyridyl) ruthenium(II) ($\text{Ru}(\text{bpy})_3^{2+}$) with tripropylamine (TPA) was reported in 1990 by Leland et al [3]. ECL has several attractive features, including absence of a background optical signal, precise control of reaction kinetics offered by controlling the applied potential, and these features make ECL a highly sensitive and selective analytical method [4-6]. $\text{Ru}(\text{bpy})_3^{2+}$ derivatives have been employed as the most important ECL labels in immunoassays and DNA bioassays [5-7]. However, they have some disadvantages, such as, high ECL potential ($>+1.0$ V, vs SCE) and low ECL efficiency (0.05 in organic solution for $\text{Ru}(\text{bpy})_3^{2+}$) [8]. Therefore, currently, intense research is under way to identify new luminophores, mechanistic studies, and analytical application.

The cyclometalated iridium(III) complexes have attracted increasing attention as promising photocatalysis materials [9, 10] and as ECL emitters for ECL assay [11-18], since they exhibit high photoluminescent efficiencies, stable redox characteristics, low potential, etc. It was found that iridium(III) complexes have high efficient PL efficiencies because both singlet and triplet excitations are contributed to PL emission and the emission color and emission efficiency can be tuned by changing main ligands and ancillary ligands [19, 20]. Since the early work of the ECL of iridium(III) complexes by the pioneering work of Nishimura [21], Wightman [22], Richter [23], and Kapturkiewicz [24], ECL works on iridium complexes are becoming hot issue in organic or aqueous solvent, which opened up the possibility in the analytical applications due to their efficient luminescence and tunable emission properties.

Much effort has been done in the design and characteristics of iridium(III) complexes to improve the ECL efficient, to low the potential and to tune the emission wavelength [25-32]. For example, many neutral or inonic iridium(III) complexes have been demonstrated to display high ECL efficiencies in organic solutions usually with a degree of efficiency much higher than $\text{Ru}(\text{bpy})_3^{2+}$. ECL efficiency of 0.88 for $(\text{pq})_2\text{Ir}(\text{pico})$, in which pq is a 2-phenylquinoline anion and pico is picolinate, was observed in the annihilation process, which is ~ 17 times higher than that of $\text{Ru}(\text{bpy})_3^{2+}$ [33]. Up to 77 and 26 times higher ECL efficiencies were observed for $(\text{pq})_2\text{Ir}(\text{acac})/\text{TPA}$ and $(\text{pq})_2\text{Ir}(\text{tmd}) \text{S}_2\text{O}_8^{2-}$ systems (tmd is 2,2,6,6-tetramethyl-3,5-heptanedione), respectively, with respect to $\text{Ru}(\text{bpy})_3^{2+}$ standard [34]. We have previously reported five iridium(III) complexes with green, orange and red ECL emission by changing the main and ancillary ligands with high efficient, in which 214-folds ECL emission is observed for $(\text{bt})_2\text{Ir}(\text{acac})$ with respect to $\text{Ru}(\text{bpy})_3^{2+}$ [35]. Although many iridium (III) complexes are known to be ECL active in a non-aqueous environment, often with a degree of efficiency much higher than $\text{Ru}(\text{bpy})_3^{2+}$, the ECL intensity of iridium (III) complexes in aqueous solution are usually lower than that in non-aqueous solvent.

In order to translate the high ECL efficiencies into a real ECL application reality, as the very first step, the emitters have to be investigated in aqueous solutions or solutions of analytical importance [36]. Considering the analytical applications of new ECL metal complexes, the contact group in the ancillary ligands as the labels should be contained. Additionally, hydrophilicity of the complexes should be considered [36, 37]. Reports about water-soluble iridium cyclometalated complexes is receiving increasing interest, in which the ancillary diimine ligands are functionalized with solubilizing groups such as sugars[38], carboxylate groups[39], as well as the

bis-cyclometalated bisqua complexes[40]. Among them, ligand with carboxylate groups has been used to synthesis novel iridium(III) complexes because the carboxylate groups provide a convenient point of attachment to other biological recognition molecules[41]. For example, a biosensor employing a novel ECL labeling reagent $[(ppy)_2Ir(dcbpy)]PF_6$ was applied in DNA detection and the sensitivity was improved more than 200 times compared to $Ru(bpy)_3^{2+}$ [42]. However, the ECL efficiency of the water-soluble iridium cyclometalated complexes is still limited. The design and synthesis of new luminophores with distinct ECL emission and high efficiency are still needed for the iridium (III) complexes in ECL analytical applications.

Our long goal is searching for water-soluble cyclometalated iridium(III) complexes with low potential and high ECL coefficient. The aim of this work is to design and characteristics of two new water-soluble cyclometalated iridium(III) complexes with hydrophilic β -acetylacetone as ancillary ligand. In this paper, two cyclometalated iridium(III) complexes, $(pq)_2Ir(sa)$ and $(pq)_2Ir(psa)$, using 2-phenylquinoline (pq) as the C^N main ligand and succinylacetone (sa) or 4,6-dioxo-6-phenylhexanoic acid (psa) as the O^O ancillary ligand are synthesized (Scheme 1). The photophysical, electrochemical and ECL properties of $(pq)_2Ir(sa)$ and $(pq)_2Ir(psa)$ in acetonitrile solvent and acetonitrile-aqueous solution are presented. A novel ECL method for the determination of water content in organic solvents using $(pq)_2Ir(sa)$ and $(pq)_2Ir(psa)$ as the ECL reagent is developed. The water-enhanced ECL double coreactant pathway was discussed.

Scheme 1

2. Experimental Section

2.1 Chemicals and Apparatus

$\text{IrCl}_3 \cdot 3\text{H}_2\text{O}$ was purchased from Shaanxi Kaida Chemical Engineering Co., Ltd. 2-phenylquinoline (pq), succinylacetone (sa), 2-phenylbenzothiazole (bt), acetylacetone (acac), potassium hexafluorophosphate (KPF_6), tris(2,2'-bipyridine) dichlororuthenium(II) hexahydrate ($\text{Ru}(\text{bpy})_3\text{Cl}_2 \cdot 6\text{H}_2\text{O}$), tetra-n-butylammonium hexafluorophosphate (TBAPF_6) and anhydrous acetonitrile (CH_3CN , 99.8%) were purchased from Sigma-Aldrich (St. Louis, MO, USA). 4,6-Dioxo-6-phenylhexanoic acid (pas) was obtained from J&K Scientific Ltd (China). 2-Methoxyethanol, trichloromethane, ethanol, methanol, n-hexane, tri-n-propylamine (TPA) were obtained from Sinopharm Chemical Reagent Co., Ltd (China). 5,6-dimethyl-1,10-phenanthroline (phen) was purchased from Tokyo Chemical Industry Co., Ltd.

The absorbance and photoluminescence (PL) spectra were recorded on a UV–Vis spectrophotometer (UV-2450, Shimadzu Corporation, Japan) and a Cary Eclipse fluorescence spectrophotometer (Varian, USA), respectively. Mass spectra (MS) were acquired in ESI positive mode using a Bruker Maxis UHR-TOF Mass Spectrometer using methanol as mobile phase.

All electrochemical experiments were carried out with a CHI 660 electrochemical workstation (Chenhua Instruments Co., Shanghai, China) and done under anhydrous conditions in an argon atmosphere glove box (Mikrouna, China). A typical three-electrode setup with a glassy carbon electrode (2.0 mm diameter), a Pt auxiliary electrode, and a quasi-reference silver wire electrode was used for electrochemical experiment unless otherwise stated. Cyclic voltammetry (CV) potentials in acetonitrile were calibrated with ferrocene as a standard, taking $E_0=0.342$ V vs. SCE.

The ECL transients and simultaneous CV and ECL measurements were recorded using a MPI-E ECL detector (Xi'an Remax Electronics, China). The ECL spectra were generated using CHI 440a and recorded using home-made ECL spectra Instrument with a Princeton Instruments PyLon100BX CCD camera (Trenton, NJ) coupled with an Acton SP2300 monochromator cooled with liquid nitrogen to -120 °C.

2.2 Synthesis of the cyclometalated iridium(III) complexes

The synthetic route of (pq)₂Ir(sa) and (pq)₂Ir(psa) is shown in Scheme 1. Two steps were employed for the synthesis of (pq)₂Ir(sa) and (pq)₂Ir(psa). Firstly, the chloride-bridged dimer [(pq)₂Ir(μ-Cl)]₂ was synthesized according to reported procedure[43]. Typically, 0.353 g IrCl₃•3H₂O (1 mmol) was added into 40 mL of 2-ethoxyethanol and water solution (V:V=3:1), which was degassed using nitrogen gas, and then 0.456g pq (2.2 mmol) was added. This mixture was refluxed for 24 h at 120 °C under nitrogen, and then naturally cooled to room temperature. The solid precipitation was all transferred to a sand core funnel for vacuum filtration. The precipitation was successively washed by water (30 mL), alcohol (30 mL), and hexane (30 mL). The crude product was dried in vacuum at 40 °C for 4 h.

Secondly, (pq)₂Ir(sa) and (pq)₂Ir(psa) were synthesized as following. Ancillary ligand sa (0.43 mmol) or pas (0.43 mmol) and triethylamine (307 μL) were added into 5.0 mL of 2-methoxyethanol containing 134.9 mg (0.106mmol) [(pq)₂Ir(μ-Cl)]₂. After degassed, the reaction mixture was stirred for 15 h at 80 °C under nitrogen. The crude product was dried in vacuum rotary. The pure product was obtained on a silica column using trichloromethane and methanol (9.5/0.5, v/v) as eluent with a yield of 50% for (pq)₂Ir(sa) and that of 57% for (pq)₂Ir(psa). The synthesis of (bt)₂Ir(acac) was carried out as reference [44] and (bt)₂Ir(phen)PF₆ was carried out as reference[33].

2.3 ECL Measurement

ECL emissions were detected with a photomultiplier tube (PMT) that was biased at -500 V unless otherwise stated. The amount of water was quantified by a increased ECL intensity ($\Delta I = I_s - I_0$), where I_s and I_0 were the ECL intensity in the presence and absence of water, respectively. ECL spectra were generated by pulsing from 0 to 80 mV past the oxidation peak of the iridium (III) complex with a step time of 0.1 s using TPA as a co-reactant.

3. Results and Discussion

3.1 Synthesis and solubility in water

At first, design idea of cyclometalated iridium(III) complexes, $(pq)_2Ir(sa)$ and $(pq)_2Ir(psa)$ should be presented. Although a number of cyclometalated iridium(III) complexes have been recently reported, regarding on the contact groups which can be used to tag proteins or other species in the reported complexes is rarely reported. In this work, pq was chosen as main ligand since it is high ECL efficiencies while sa or psa was chosen as ancillary ligand since they contain β -acetylacetone and $-COOH$. The latter could be used to tag proteins or other species with primary amine groups.

The synthetic procedure for the cyclometalated iridium(III) complexes consisted of two steps, as described in Experimental Section. The structures of $(pq)_2Ir(sa)$, $(pq)_2Ir(psa)$, $(bt)_2Ir(acac)$ and $(bt)_2Ir(phen)PF_6$ were shown in Figure S-1. The chemical structures of $(pq)_2Ir(sa)$ and $(pq)_2Ir(psa)$ synthesized were further confirmed by ESI-MS (Figure S2, S3 in the Supporting Information). The ESI mass spectrum features a major peak centered at m/z 757.1681 corresponding to $(pq)_2Ir(sa)$ (calculated $[M-H]^-$ 757.1686), while that centered at m/z 819.1827 (calculated $[M-H]^-$ 819.1843) corresponding to $(pq)_2Ir(psa)$. For control experiment, $(bt)_2Ir(acac)$ and $(bt)_2Ir(phen)^+$ show features peak at 713.0895 ($[M+H]^+$) and 821.1369 ($[M]^+$),

respectively (Figure S4, S5 in the Supporting Information). The data demonstrate that the target complexes are successfully synthesized.

For the potential use in analytical application, the solubility of the synthesized complexes as ECL labels in water is very important. Therefore, the solubility of $(pq)_2Ir(psa)$ and $(pq)_2Ir(sa)$ in 5:95 (v/v) CH_3CN/H_2O was checked using UV-Vis absorption method. Molar absorption coefficient is an intrinsic property of the compound in the solution, and it should not be changed with the concentration of the compound unless the concentration is too high or the complex is not stable, or the complex is not soluble under the experimental conditions. The results showed that the absorbability was not changed for concentration of $(pq)_2Ir(psa)$ down to 10 μM and for $(pq)_2Ir(sa)$ down to 0.1 mM (See Figure S-6). The solubility of two synthesized complexes is 10 μM and 0.1 mM for $(pq)_2Ir(psa)$ and $(pq)_2Ir(sa)$, respectively. This indicates that the solubility of $(pq)_2Ir(sa)$ in 5:95 (v/v) CH_3CN/H_2O is higher than that of $(pq)_2Ir(psa)$ in the same solvent. The lower solubility of $(pq)_2Ir(psa)$ is attributed to the fact the cyclometalating ligand in $(pq)_2Ir(psa)$ contains a hydrophobic phenyl group. Importantly, the solubility of the synthesized complexes is much larger than that of typical $Ir(ppy)_3^{2+}$ ($\sim 0.1 \mu M$ in 5:95 (v/v) CH_3CN/H_2O) [23], attributed to hydrophilic groups, $-COOH$ and β -acetylacetone in ancillary ligand. A satisfactory solubility of two cyclometalated iridium(III) complexes synthesized in this work is, therefore, evident.

3.2 Photophysical and electrochemical properties of $(pq)_2Ir(sa)$ and $(pq)_2Ir(psa)$ in acetonitrile

Photophysical and electrochemical properties of $(pq)_2Ir(sa)$ and $(pq)_2Ir(psa)$ was studied in acetonitrile so that they was prepared with reported data of other complex. Figure 1 shows the UV-Vis absorption spectra of $(pq)_2Ir(sa)$ and $(pq)_2Ir(psa)$

in acetonitrile solvent under air-saturated conditions. The correspondingly spectroscopic data of (pq)₂Ir(sa) and (pq)₂Ir(psa) are summarized in Table 1. It can be seen that (pq)₂Ir(sa) and (pq)₂Ir(psa) have strong intra-ligand absorption bands (π - π^*) in the UV region (266 nm for (pq)₂Ir(sa), 267 nm for (pq)₂Ir(psa)), as typically observed for the cyclometalated iridium complex [45], and absorption bands at 339 nm for (pq)₂Ir(sa) and at 340 nm for (pq)₂Ir(psa), assigned to the metal-to-ligand charge transfer transitions (MLCT). These UV–Vis absorption spectra are similar to that of (pq)₂Ir(acac) in which common cyclometalating ligand 2-phenylquinoline is used as the C[^]N main ligand while β -acetylacetone (acac) is used as the O[^]O ancillary ligand [44]. The molar extinction coefficient (ϵ) of (pq)₂Ir(psa) ($\epsilon_{267}=6.7\times10^4$ M⁻¹cm⁻¹, $\epsilon_{340}=2.7\times10^4$ M⁻¹cm⁻¹) in air-saturated acetonitrile is slightly higher than that of (pq)₂Ir(sa) ($\epsilon_{266}=5.3\times10^4$ M⁻¹cm⁻¹, $\epsilon_{339}=2.1\times10^4$ M⁻¹cm⁻¹), attributed to an increasing conjugation from the phenyl group in psa. The molar extinction coefficient of (pq)₂Ir(sa) in air-saturated acetonitrile is slightly lower than that of (pq)₂Ir(acac), attributed to the electron-attracting group, carboxyl group in sa [37].

Figure 1

Figure 2 shows photoluminescence (PL) emission spectra of (pq)₂Ir(sa) and (pq)₂Ir(psa) in air- saturated acetonitrile and N₂-saturated acetonitrile, respectively. In both cases, two complexes all display structureless orange emission band with a maximum wavelength at 608 nm for (pq)₂Ir(sa) and at 601 nm for (pq)₂Ir(psa). PL efficiency was calculated to be 0.0365 for (pq)₂Ir(sa) and 0.0718 for (pq)₂Ir(psa) in air-saturated acetonitrile when Ru(bpy)₃Cl₂ was used as the standard ($\Phi_{\text{PL}}=0.059$ in CH₃CN).³¹ Compared with PL emission wavelength of (pq)₂Ir(acac) ($\lambda_{\text{em}}=597$ nm) [37], replacement of acac with sa or psa as ancillary ligand resulted in red-shifts of 11 nm for (pq)₂Ir(sa) and 4 nm for (pq)₂Ir(psa) at PL emission wavelengths. The PL

emission intensity of two iridium (III) complexes in N₂-saturated acetonitrile is higher than that in air-saturated acetonitrile. This indicates that the dissolved oxygen in acetonitrile can quench the PL emission. This suggests that these two iridium (III) complexes will be potentially applied to develop a PL method for the determination of oxygen.

Figure 2

Figure 3 shows cyclic voltammograms of (pq)₂Ir(sa) (1.0 mM, dot line) and (pq)₂Ir(psa) (1.0 mM, solid line) in CH₃CN containing 0.10 M TBAPF₆. As shown in oxidation range of Figure 3, a quasi-reversible oxidation wave of (pq)₂Ir(sa) and (pq)₂Ir(psa) appears at +0.79V and +0.8 V vs SCE, respectively. The oxidation potential of (pq)₂Ir(psa) is slightly more positive than that of (pq)₂Ir(sa), attributed to the fact that the ancillary ligand (psa) makes central metal be difficultly oxidized since the steric-hindrance effect from a big phenyl group in psa is larger than that from a small methyl group in sa. The less reversibility of these two complexes may be explained by the orbitals that receive contributions from M–C–σ bonds between Ir-d orbitals and pyridine orbitals [47]. This oxidation peak potential is more negative than that of (pq)₂Ir(acac) appeared at +0.99 V vs. SCE in acetonitrile [34]. Compared with Ru(bpy)₃²⁺ (formal potential of 1.1 V vs SCE in air-saturated acetonitrile) [48], these two cyclometalated iridium(III) complexes have a lower oxidation potential. As shown in reduction range of Figure 3, a quasi-reversible reduction wave of (pq)₂Ir(sa) appears at about -1.86 V vs SCE while irreversible reduction wave of (pq)₂Ir(psa) appears at about -1.88 V vs SCE. The reduction potential of (pq)₂Ir(psa) is slightly more negative than that of (pq)₂Ir(sa), attributed to the steric-hindrance effect, that is to say, the fact that a big phenyl group in psa is larger than that from a small methyl group in sa, resulted the ancillary ligand (psa) itself difficultly to be reduced. The

reduction peak potentials of (pq)₂Ir(sa) and (pq)₂Ir(psa) are more positive than that of *fac*-Ir(ppy)₃ (-2.2 V vs. SCE) and are more negative than that of (pq)₂Ir(acac) (-1.63 V vs. SCE) [34] and that of Ru(bpy)₃²⁺ (-1.33 V vs. SCE) in acetonitrile. These are mainly attributed to the electron-attracting group in (pq)₂Ir(sa) and (pq)₂Ir(psa). A satisfactory lower oxidation potential of two cyclometalated iridium(III) complexes synthesized in this work is, therefore, evident, and this greatly benefit the coreactant ECL process for ECL analysis.

Figure 3

3.3 ECL properties in N₂-saturated acetonitrile containing TPA

There are usually two mechanistic processes in ECL, including ion annihilation processes and coreactant processes. In ion annihilation, electrolytic generation of both the oxidized and reduced ECL precursors is required. The absolute ECL yields obtained by ion annihilation in acetonitrile containing 0.10 M TBAPF₆ when Ru(bpy)₃²⁺ was as a standard ($\Phi=0.05$ in MeCN) was calculated to be 0.30 for (pq)₂Ir(sa) and 0.51 for (pq)₂Ir(psa) [49]. In coreactant processes, TPA [3] was used as reductive coreactant to generate ECL. Figure 4 A shows ECL intensity-potential curves of (pq)₂Ir(sa) and (pq)₂Ir(psa) at a platinum electrode in N₂-saturated acetonitrile containing 0.10 M TBAPF₆ and 25 mM TPA. One ECL peak at +0.91 V was observed for 1 mM (pq)₂Ir(psa) in the presence of 10 mM TPA, which is attributed to the classic coreactant mechanism [50], that is, the radical TPA (formed during TPA oxidation) reduces (pq)₂Ir⁺(psa) (formed from the oxidation of (pq)₂Ir(psa)) to produce the excited state (pq)₂Ir^{*}(psa), resulted in generating ECL emission. One ECL emission at +0.87 V was observed for (pq)₂Ir(sa), followed by the same ECL coreactant mechanism as above. The ECL peak potential of (pq)₂Ir(psa) is more positive than that of (pq)₂Ir(sa), attributed to a higher oxidation potential of

(pq)₂Ir(psa). The ECL peak intensity of (pq)₂Ir(psa) is higher than that of (pq)₂Ir(sa). This is possibly attributed to the fact that (pq)₂Ir(psa) has a high PL efficiency.

Figure 4

The ECL spectra of (pq)₂Ir(sa) and (pq)₂Ir(psa) were obtained by pulsing between 0 and 80 mV past the oxidation peak potential in the presence of TPA with a pulse time of 0.1 s, respectively. As shown in Figure 4B, (pq)₂Ir(sa) and (pq)₂Ir(psa) produced bright orange ECL emission in oxidative-reduction coreactant. The maximum ECL emissions of (pq)₂Ir(sa) and (pq)₂Ir(psa) were 621 nm and 610 nm, respectively. The slightly red-shift in the ECL spectra was observed, compared with that of PL emission. This is maybe ascribed to the different concentrations used in the PL measurement (μ M level) and ECL measurement (mM level) and the difference instruments with different resolutions and slit widths used for PL and ECL measurements [35,51]. The similar ECL and PL emission indicate that the excited state is similar in the PL emission and the ECL coreactant emission. ECL efficiency of (pq)₂Ir(sa)/TPA and (pq)₂Ir(psa)/TPA is calculated when standard Ru(bpy)₃²⁺/TPA was used as the standard (ECL efficiency is 100%) under same conditions. The ECL efficiencies for (pq)₂Ir(sa)/TPA and (pq)₂Ir(psa)/TPA are 21.3 and 35.5-folds higher than that of Ru(bpy)₃²⁺/TPA system in acetonitrile solvent, respectively. They are also much higher than that of Ir(ppy)₃²⁺/TPA ($\Phi=0.33$) [23]. A satisfactory ECL efficiencies of two cyclometalated iridium(III) complexes synthesized in this work are, therefore, obtained, and this greatly benefits the ECL analysis.

3.4 ECL properties in air-saturated acetonitrile containing TPA and water

Although many iridium (III) complexes are known to be ECL active in a non-aqueous environment, often with a degree of efficiency much higher than Ru(bpy)₃²⁺, the ECL intensity of iridium (III) complexes in aqueous solution are

usually lower than that in non-aqueous solvent. Qijun Song group recently reported that hydroxide and ethoxide ions can enhance the ECL intensity of (pq)₂Ir(acac) in organic solvents without TPA [52]. They demonstrated that trace water in the organic solvent could undergo dissociation in the presence of alkali reagent and produce hydroxide ions and the hydroxide ion could act as the coreactant for the available ECL system. We found that in air-saturated acetonitrile containing TPA, a small amount of water can greatly increase the ECL intensity of (pq)₂Ir(psa) and (pq)₂Ir(sa) in neutral aqueous solution. Therefore, we investigated ECL behaviors of (pq)₂Ir(psa) and (pq)₂Ir(sa) in air-saturated acetonitrile in the absence/presence of TPA with addition of trace water.

Figure 5 shows ECL intensity-potential curves of (pq)₂Ir(sa) and (pq)₂Ir(psa) in air-saturated acetonitrile containing 0.10 M TBAPF₆ and 25 mM TPA with addition of different amounts of water. Upon addition of water to a solution of (pq)₂Ir(psa) and (pq)₂Ir(sa), an increase in ECL intensity was observed. The ECL peak intensity of (pq)₂Ir(psa) directly increases with an increase of the concentration of water in CH₃CN from 0.02% to 4.0% (v/v) (Figure 5A). The regression equation was $\Delta I_{\text{ECL}} = 2.1 \times 10^5 C + 492$ (where C is the water content in volume ratio) with the correlation coefficient of 0.9982. The ECL peak intensity of (pq)₂Ir(sa) directly increases with an increase of the concentration of water in CH₃CN from 0.05% to 4.0% (v/v) (Figure 5B). The regression equation was $\Delta I_{\text{ECL}} = 1.59 \times 10^5 C + 64$ with the correlation coefficient of 0.9937 (where c is the water content in volume ratio). The sensitivity (the slope) of the linear calibration curve from (pq)₂Ir(psa) is higher than that of (pq)₂Ir(sa). The detection limit is calculated to be 0.01% for (pq)₂Ir(psa) and 0.03% for (pq)₂Ir(sa), respectively. The two as-prepared complexes show different sensitivities and detection limits towards water determination, attributed to the fact

that the relative ECL efficiency of (pq)₂Ir(psa) is higher than that of (pq)₂Ir(sa). It should be noted that trace amounts of water from air will increase the background ECL intensity when the experiment is conducted in air-saturated conditions. To lower the detection limit, the tested solution in ECL cell should be saturated by the air from water-clear silicon column.

The ECL efficiencies for (pq)₂Ir(sa)/TPA and (pq)₂Ir(psa)/TPA in the mixed CH₃CN:H₂O (v/v=50:50) are 1.89 and 2.35-folds higher than that of Ru(bpy)₃²⁺/TPA system in the same condition, respectively. And they are also much higher than that of Ir(ppy)₃²⁺ in mixed solution ($\Phi=0.0044$) [23]. Therefore, the developed ECL method using (pq)₂Ir(psa) or (pq)₂Ir(sa) will be potentially applied for the determination of trace water in organic solvents.

Figure 5

ECL intensity–potential curves of 2.00 μ M (pq)₂Ir(psa) in air-saturated acetonitrile in the absence of TPA after addition of different amounts of water was recorded. The results showed that the ECL intensity increased with increasing an amount of water from 0.5%-3.0% (v/v) (Figure S-7). The regression equation was $\Delta I_{\text{ECL}} = 2.3 \times 10^4 C + 22$ (where C is the water content in volume ratio) with the correlation coefficient of 0.9862. The detection limit is 0.1% (v/v). The sensitivity of determination of water using (pq)₂Ir(psa) in the presence of TPA is 9.1 folds higher than that in the absence of TPA. This indicates that TPA enhance the ECL intensity of (pq)₂Ir(psa) in the presence of water.

3.5 Exploration of the water-enhanced ECL pathway

In order to preliminarily understand the water-enhanced ECL pathway of (pq)₂Ir(sa) and (pq)₂Ir(psa) in acetonitrile containing TPA, the following experiments were conducted. Firstly, the ECL behaviors of other two cyclometalated iridium(III)

complexes, $(bt)_2Ir(acac)$ and $(bt)_2Ir(phen)^+$, as showed in Figure S-1, were studied in air-saturated acetonitrile containing 0.10 M TBAPF₆ and 25 mM TPA (Figure 6). The results showed that the ECL peak intensity of $(bt)_2Ir(acac)$ increases, like $(pq)_2Ir(sa)$ and $(pq)_2Ir(psa)$ case, while the ECL peak intensity of $(bt)_2Ir(phen)$ was not changed after addition of water into the acetonitrile solution. This is attributed to Keto-Enol tautomerism of acetylacetone and the derived β -acetylacetone as ancillary ligand. This indicates that ancillary ligand with β -acetylacetone is key issue for water-enhanced ECL.

Figure 6

Secondly, effect of other hydrophilic compounds, such as, methanol and ethanol, on the ECL intensity of $(pq)_2Ir(sa)$ and $(pq)_2Ir(psa)$ in air-saturated acetonitrile containing TPA was studied. We found that small amount of methanol/ethanol can also increase the ECL peak intensity of $(pq)_2Ir(psa)$ and $(pq)_2Ir(sa)$ in air-saturated acetonitrile containing TPA (see Figure 7). These indicates that the tested compounds contained -OH group, like water, can also be used as a coreactant substance for ECL of $(pq)_2Ir(sa)/TPA$ and $(pq)_2Ir(psa)/TPA$. The sensitivity of determination of water (1.59×10^5) using $(pq)_2Ir(sa)$ in the presence of TPA is 5.9 and 15.9 folds higher than that for methanol (2.7×10^4) and ethanol(1.0×10^4), respectively.

Figure 7

Thirdly, a chemiluminescence experiment for $(pq)_2Ir(psa)$ using solid lead dioxide as oxidizing agent and 0.1 M NaOH as reducing agent was performed [53]. 0.3 mg $(pq)_2Ir(psa)$ was dissolved in 0.2 mL acetonitrile and then diluted by 0.05 M H₂SO₄ into 4 mL to form a 0.1mM $(pq)_2Ir(psa)$ solution. This $(pq)_2Ir(psa)$ solution was treated with solid lead dioxide (0.32g). When 3 mL of the resulted oxidized

(pq)₂Ir(psa) solution was mixed with 0.2 mL of 0.1 M NaOH, an obvious CL light emission was observed. Compared with no oxidization of (pq)₂Ir(psa) by PbO₂, the intensity after oxidation by PbO₂ was higher than that without oxidization by PbO₂ (Figure S-8). The result indicates that the oxidized (pq)₂Ir⁺(psa) generated by the oxidation of PbO₂ can reduce OH⁻ to form radical OH[·], which then reduce (pq)₂Ir⁺(psa) to form excited (pq)₂Ir^{*}(psa) [53].

Based on data obtained above and typical coreactant ECL pathway[50], a double coreactant ECL pathway was proposed for the two complexes in acetonitrile containing TPA and water as following using (pq)₂Ir(psa) as a model complex.

Brifly, (pq)₂Ir(psa) and TPA were electrochemically oxidized to (pq)₂Ir⁺(psa) (eq.1) and TPA^{·+} (eq.2), respectively. And then the chemiluminescence reactions carry out in two ways. One way is that TPA^{·+} deprotonates to radical TPA[·] (eq.3) and then radical TPA[·] reacts with (pq)₂Ir⁺(psa) to form excited (pq)₂Ir^{*}(psa) (eq.4) , and finally (pq)₂Ir^{*}(psa) goes down to a ground state to produce ECL emission (eq.7). Another way is that the oxidized (pq)₂Ir⁺(psa) reacts with OH⁻ in water to produce radical OH[·](eq.5), and then (pq)₂Ir^{·+}(psa) reacts with OH[·] to form excited (pq)₂Ir^{*}(psa) (eq. 6), and finally generates ECL emission (eq.7).

4. Conclusions

The two cyclometalated iridium(III) complexes, (pq)₂Ir(sa) and (pq)₂Ir(psa), with lower potential and high ECL coefficient, were investigated at first time. The solubility was reached down to 0.1 mM for (pq)₂Ir(sa) and 10 μM for (pq)₂Ir(psa) in acetonitrile-water (v/v=5:95) solution. The oxidation peak potential of (pq)₂Ir(sa) and (pq)₂Ir(psa) appears at +0.79V and +0.8 V vs SCE, respectively. The ECL efficiencies of (pq)₂Ir(sa)/TPA and (pq)₂Ir(psa)/TPA were calculated to be 21.3 and 35.5-folds higher than that of Ru(bpy)₃²⁺/TPA in acetonitrile solvent. The ECL methods have been developed for the determination of trace water in organic solvents with the detection limit of 0.01% by using (pq)₂Ir(sa) and 0.03% by using (pq)₂Ir(psa). Additionally, the water-enhanced ECL pathway in the presence of TPA is proposed as a double coreactant ECL pathway. This work demonstrates that introduction of -COOH and phenyl group into β-acetylacetone as the ancillary ligand of cyclometalated iridium(III) complexes resulted in good solubility in water, lower oxidation potential and higher ECL efficiency compared with that other complex previously reported in organic solvent. These two cyclometalated iridium(III) complexes are promising to be used as ECL emitter in analytical application.

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Scheme, Figures and Captions

Scheme 1. The synthesis procedure of the iridium complexes

Figure 1. UV-vis absorption spectra of (pq)₂Ir(sa) (1.00×10^{-5} M, dot line) and (pq)₂Ir(psa) (1.00×10^{-5} M, solid line) in air-saturated acetonitrile.

Figure 2. Photoluminescence emission spectra of (pq)₂Ir(sa) (1.00×10^{-5} M, dot line) and (pq)₂Ir(psa) (1.00×10^{-5} M, solid line) in acetonitrile. $\lambda_{\text{ex}}=365$ nm. (A) Air-saturated acetonitrile; (B) N₂-saturated acetonitrile.

Figure 3. Cyclic voltammograms of (pq)₂Ir(sa) (1.0 mM, dash line) and (pq)₂Ir(psa) (1.0 mM, solid line) at Pt electrode in nitrogen-saturated acetonitrile containing 0.10 M TBAPF₆, scan rate 50 mV/s.

Figure 4. (A) ECL intensity-potential curves and normalized ECL spectra of (pq)₂Ir(sa) (1.0 mM, dot line) and (pq)₂Ir(psa) (1.0 mM, solid line) in N₂-saturated acetonitrile containing 0.10 M TBAPF₆ and 10 mM TPA at Pt electrode (2.0 mm diameter). (A) scan rate 50 mV/s; (B) ECL spectrum obtained by stepping from 0 to 1.0 V with a pulse width of 0.1 s.

Figure 5. (A) ECL intensity –potential curves of 2.00 μ M (pq)₂Ir(psa) in air-saturated acetonitrile containing 0.10 M TBAPF₆ and 25 mM TPA with addition of different amounts of water at a glassy carbon electrode. The water contents (v/v) from a to i are 0, 0.02, 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 4.0(%), respectively. Scan rate: 50 mV/s. (B) ECL intensity–potential curves of 5.00 μ M (pq)₂Ir(sa) in air-saturated acetonitrile containing 0.10 M TBAPF₆ and 25 mM TPA with addition of different amounts of water, at a glassy carbon electrode. The water contents (v/v) from a to f are 0, 0.05, 0.5, 1.0, 1.5, 2.5, 4.0(%), respectively. Scan rate: 50 mV/s. Inset: The linear relationships between the water contents and the ECL intensities (ΔI) in acetonitrile.

Figure 6. ECL intensity-potential curves of 1 μM $(\text{bt})_2\text{Ir}(\text{acac})$ (A) and 10 μM $(\text{bt})_2\text{Ir}(\text{phen})$ (B) in air-saturated acetonitrile containing 0.10 M TBAPF_6 and 25 mM TPA in the absence (a) and presence of 0.5% (b) and 1.0% (c) water. Scan rate: 50 mV/s.

Figure 7. ECL intensity-potential curves of 2.00 μM $(\text{pq})_2\text{Ir}(\text{psa})$ in air-saturated acetonitrile containing 0.10 M TBAPF_6 and 25 mM TPA in the absence (a) and presence of 0.5% (b) and 1.0 % (c) methanol (A) and ethanol (B). Scan rate: 50 mV/s.

Table 1. Spectroscopic and electrochemical data of (pq)₂Ir(sa) and (pq)₂Ir(psa) in acetonitrile.

Complex	$\lambda_{\text{max}}[\text{nm}]$	$\epsilon(10^4\text{M}^{-1}\text{cm}^{-1})$	$\lambda_{\text{em}}[\text{nm}]$	Φ_{PL}	air-sat	$E_{\text{ox}}^{[\text{a}]}$	$E_{\text{red}}^{[\text{a}]}$
(pq) ₂ Ir(sa)	266,339	5.30, 2.10	608	0.0365		+0.79V	-1.86V
(pq) ₂ Ir(psa)	267,340	6.70, 2.70	601	0.0718		+0.80V	-1.88V

^[a] All potentials were referenced to SCE using ferrocene as an internal standard ($E^0 = 0.342$ V vs SCE) and obtained by square wave voltammetry technique.

Main ligand

pq

Ancillary ligand (o-o)

sa

psa

Figureg

Figure 1

Figure 2

Figure 3

Figure 4

Figure 5

Figure 6

Figure 7