Light-induced catalytic and cytotoxic properties of phosphorescent transition metal compounds with a d⁸ electronic configuration

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Transition metal compounds are well documented to have diverse applications such as in catalysis, light-emitting materials and therapeutics. In the areas of photocatalysis and photodynamic therapy, metal compounds of heavy transition metals are highly sought after because they can give rise to triplet excited states upon photoexcitation. The long lifetimes (more than 1 μs) of the triplet states of transition metal compounds allow for bimolecular reactions/processes such as energy transfer and/or electron transfer to occur. Reactions of triplet excited states of luminescent metal compounds with oxygen in cells may generate reactive oxygen species and/or induce damage to DNA, leading to cell death. This article recaps the recent findings on photochemical and phototoxic properties of luminescent platinum(II) and gold(III) compounds both from the literature and experimental results from our group.

1. Introduction

Metal compounds have long been demonstrated to have diverse applications in catalysis [1,2], therapeutics usages [3,4] and also as light-emitting materials [5,6]. In the face of the ever-growing demand for energy...
and limited reserves of natural energy sources, research in the conversion of light energy to chemical energy has been rapidly expanding over the past decades. In this endeavour, there is a surge of interest to develop new metal compounds as ‘energy converters’ because of their intrinsic ability to absorb light energy in the UV–visible spectral region. Upon photoirradiation, a metal compound can be excited to a singlet state with an electron populated in an energetically higher-lying orbital. The singlet state, in the presence of a metal ion such as that of ruthenium, platinum, gold or iridium, can decay through intersystem crossing (ISC) via spin–orbit coupling to a low-lying triplet excited state. Since the decay of triplet excited state to singlet ground state is a spin-forbidden process, the triplet excited states of transition metal compounds commonly have a lifetime in the microsecond range, which is long-lived enough to allow for bimolecular reactions/processes such as energy transfer and/or electron transfer to occur. Perhaps the most extensively studied class of metal compounds in this regard is tris(2,2′-bipyridine)ruthenium(II) ([Ru(bpy)₃]²⁺) and its derivatives [7]. The wide scope of photochemistry demonstrated by [Ru(bpy)₃]²⁺ has subsequently stimulated immense research activities on the photophysical and photochemical properties of other transition metal compounds including those of platinum [8], iridium [9], osmium [10,11] and more recently gold [12].

As technology advances, interdisciplinary research such as photocatalysis and photodynamic therapy (PDT) using phosphorescent transition metal compounds emerges. Photocatalysis takes advantage of harnessing light energy for driving chemical reactions. A photocatalyst in a reaction mixture could be excited by light at specific wavelengths so that other substrate(s) present would be least affected by the light irradiation.

PDT involves the activation, by light at a specific wavelength, of an administrated photosensitizer to undergo energy transfer and/or electron transfer in biological systems such as generation of the highly cytotoxic singlet oxygen ¹O₂ [13]. When the photosensitizer absorbs light energy, it will be excited to a singlet state (¹PS*) and subsequently to a triplet state (³PS*) after ISC. The triplet state has a relatively long lifetime to transfer its energy to the oxygen molecules present in the biological system to form reactive oxygen species which can damage cells/tissues through oxidative processes (figure 1) [14]. An effective photosensitizer for PDT would have (i) preferential localization in the targeted area (e.g. cancer tissue), (ii) strong absorbance at longer wavelengths (i.e. more than 500 nm) for achieving a better penetrating power in the biological tissues, (iii) high quantum yield and long lifetime of the triplet excited state, and (iv) non-toxicity under dark conditions.

Haematoporphyrin, with a commercial name of Photofrin, was first approved in Canada and subsequently approved by the US Food and Drug Administration to be used clinically as a PDT photosensitizer for the treatment of some types of cancers. Other PDT photosensitizers including metalloporphyrin compounds with strong absorbance at 650–850 nm have also been identified (figure 2) [15].
Figure 2. Selected examples of effective photosensitizers for photodynamic therapy.

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\text{Pt}_2^* + RX \rightarrow \text{Pt}_2X + R \quad (X = \text{Cl, Br})
\]

Figure 3. Examples of atom abstraction reactions by Pt\(_2^*\). Light source: 200 W mercury lamp, \(\lambda > 335\) nm.

Conventional PDT requires oxygen as reactive species to kill cancer cells. Yet, most tumour tissues are under hypoxic conditions thereby limiting the scope of applications of this PDT. Recent works by Sadler and co-workers [16,17] and Schatzschneider [18] revealed that light-induced chemical reduction or photoassisted bond dissociation could be used to convert a metal compound to a highly active form for cancer treatment. This photochemical activation has been demonstrated to circumvent the problem of oxygen deficiency. Metal compounds intrinsically have the capability to absorb visible light more efficiently than organic compounds. Moreover, the large variety of metals and ligands allows systematic tuning of the photochemical properties of metal compounds to achieve specific applications. In the following sections, we present a concise discussion on recent developments of photoinduced catalytic and cytotoxic properties of some platinum(II) and gold(III) compounds.

2. Photoinduced catalytic properties of platinum(II) compounds

While the clinical success of cisplatin is an important driving force for the development of platinum(II) chemistry, extensive studies have also been directed to study the photochemistry and material applications of luminescent platinum(II) compounds [5,6]. Luminescent platinum(II) compounds are well documented to display spectacular photophysical and photochemical properties. The fundamental difference between platinum(II) and other transition metals (e.g. ruthenium(II), iridium(III)) is that platinum(II) has a square planar coordination geometry leaving two axial sites vacant. Such a coordination geometry allows for inner-sphere electron transfer, metal–substrate interaction(s) and Pt···Pt interaction(s) to take place at the platinum(II) site. Because of the heavy-atom effect, platinum(II) compounds could attain a triplet state in most cases upon photoexcitation. As the relaxation from the triplet excited state to the singlet ground state is a spin-forbidden process, the excited platinum(II) compound usually has a sufficient lifetime to participate in bimolecular reaction(s) in solution.
A most thoroughly studied platinum(II) compound showing remarkable excited-state chemistry is the diplatinum(II) compound \([\text{Pt}_2(\mu-\text{P}_2\text{O}_5\text{H}_2)_4]^{4-}\) (abbreviated Pt\(_2\)) [19]. The two Pt\(^{2+}\) ions in this compound are separated by 2.925 Å, indicative of significant Pt···Pt interaction. The low-energy triplet excited state of \([\text{Pt}_2(\mu-\text{P}_2\text{O}_5\text{H}_2)_4]^{4-}\) is assigned to come from the 5d\(\sigma^*\) \(\rightarrow\) 6p\(\sigma\) transition. In the \(3[5d\sigma^*6p\sigma]\) excited state, \(3\text{Pt}_2^*\) is both a strong reductant and also a strong oxidant. It is capable of reducing bipyridinium acceptors and meanwhile oxidizing a series of amines. The uniqueness of the photochemical properties of \(3\text{Pt}_2^*\) is also revealed by its ability to undergo excited-state atom transfer (figure 3). \(3\text{Pt}_2^*\) can abstract halogen atom from alkyl and aryl halides to form Pt\(_2\)X species (wherein X = halide). Pt\(_2^*\) is capable of cleaving C–H bonds through hydrogen atom abstraction. For instance, in the presence of a catalytic amount of \([\text{Pt}_2(\mu-\text{P}_2\text{O}_5\text{H}_2)_4]^{4-}\), isopropyl alcohol can be photochemically converted to acetone and hydrogen. Other photochemical reactions include the light-induced cleavage of allylic C–H bonds of cycloalkenes in the presence of \([\text{Pt}_2(\mu-\text{P}_2\text{O}_5\text{H}_2)_4]^{4-}\).

In recent years, research on platinum(II) photochemistry is mainly focused on hydrogen production and singlet oxygen generation. Luminescent platinum(II) compounds containing both terpyridine and acetylide ligands have been reported to effectively catalyse hydrogen production from Hantzsch dihydropyridines in acetonitrile [20]. In 2004, Tung and co-workers employed a series of platinum(II) compounds supported by terpyridine and acetylide ligands to photocatalyse hydrogen production from Hantzsch dihydropyridine derivatives in quantitative yields with up to 1000 turnovers. The mechanism of the photochemical reaction was suggested to involve the bi-radical character of the triplet excited state, in which hydrogen abstraction from the N-bonded hydrogen atom occurs. Substrates with N-alkyl units were found to undergo photocatalytic dealkylation (figure 4).

In 2006, Eisenberg and co-workers reported using a similar platinum(II) compound (PtTA, figure 5) as photosensitizer for light-induced hydrogen production from an acetonitrile/water mixture [21]. Subsequent quenching studies of the excited state of PtTA using various dialkylated bipyridinium cations and triethanolamine (TEOA) revealed that the quenching rate constants...
Figure 5. The platinum(II) compound PtTA and dialkylated bipyridinium cations examined by Eisenberg and co-workers.

Figure 6. Platinum(II) quaterpyridine compound to be incorporated in Nafion membrane.

are close to the diffusion-controlled limit [22]. Upon photoirradiation of a mixture of the PtTA, methyl viologen (MV$^{2+}$), TEOA, and colloidal Pt in an acetonitrile/water solution, the blue colour characteristic of MV$^{+•}$ radical developed and hydrogen with turnover number as high as 800 was obtained after 20 h (light source: 200 W mercury xenon lamp, $\lambda > 410$ nm).

Eisenberg and co-workers in 2009 reported a study on PtTA and TEOA in the presence of a cobalt(III) compound cobaloxime [Co(dmgH)$_2$(py)Cl] (dmgH$_2$ = dimethylglyoxime) for photoinduced hydrogen generation in an aqueous medium [23]. The initial step of the reaction can be either oxidative quenching or reductive quenching of the triplet excited state of PtTA by cobaloxime or TEOA to generate cobalt(II) and TEOA$^{+}$, respectively. Cobalt(II) was further reduced to cobalt(I) which reacted with proton to give a cobalt(III) hydride. Subsequent reduction of cobalt(III) hydride and protonation gave molecular hydrogen with regeneration of cobalt(II). Under the optimized conditions, more than 2100 turnovers of hydrogen were obtained in 10 h of photoirradiation (light source: 200 W mercury xenon lamp, $\lambda > 410$ nm).

Apart from the usage in hydrogen production, a number of luminescent platinum(II) compounds have been shown to be excellent photosensitizers for singlet oxygen generation. In 2001, Che and co-workers reported the use of a platinum(II) compound supported by quaterpyridine ligand incorporated in Nafion membrane to catalyse the oxidation of alkenes such as trans-stilbene (figure 6) [24]. Generation of singlet oxygen was confirmed by the formation of nitroxide free radical of 2,2,6,6-tetramethylpiperidine. Notably, the Nafion-supported platinum(II) photocatalyst could be recovered easily and used repeatedly without significant loss in activity. Tung and co-workers [25] subsequently designed four different kinds of polymer-supported platinum(II) photocatalyst (figure 7). Mesoporous molecular sieves, IRA-200 resin [26], silica and polymer matrices [27] incorporated with platinum(II) compounds were found to be efficient photosensitizers for production of singlet oxygen for catalysing alkene oxidation (figure 8).
Figure 7. Platinum(II) compounds incorporated on various types of solid support.

Figure 8. Photoinduced aerobic oxidation of olefins catalysed by supported platinum(II) compounds reported by Tung and co-workers. Light source: 500 W high-pressure Hanovia mercury lamp, $\lambda > 450$ nm.

In 2010, Che and co-workers examined the photophysical and photochemical properties of two cyclometalated platinum(II) compounds (Pt1 and Pt2, figure 9) [28]. Compound Pt1 is highly emissive with emission maxima at 556–562 nm in both CH$_3$CN and CH$_2$Cl$_2$ solutions. Its emission could be readily quenched by MV$^{2+}$ following Stern–Volmer behaviour at a rate constant of $6.3 \times 10^9$ M$^{-1}$ s$^{-1}$. Compound Pt2 is non-emissive in CH$_3$CN, and this is rationalized by the presence of viologen acceptor on the ligand, leading to a rapid quenching of the excited state. By ultrafast time-resolved absorption measurements, upon photoexcitation, the intramolecular reduction of the appended MV$^{2+}$ by the excited Pt$^{II}(C^\wedge N^\wedge N)$ chromophore occurs. Net formation
of MV⁺• radical can be observed when TEOA is added. Upon photoirradiation of a mixture of Pt2 and TEOA in degassed CH3CN, the solution colour changed from pale yellow to blue-green. The development of absorption bands at 395 and 605 nm indicated the formation of the MV⁺• radical. Femtosecond time-resolved absorption spectral measurement clearly showed that the formation of the charge-separated species ClPt III(C∧N∧N-PhMV⁺•) is faster than the back-electron transfer by three orders of magnitude. Compound Pt2 had also been examined for photoinduced hydrogen production. Upon photoirradiation of a degassed mixture of Pt2 (2 × 10⁻⁵ M), TEOA (0.1 M) and colloidal Pt (5 × 10⁻⁵ M) with visible light (300 W xenon lamp, λ > 400 nm), the blue colour characteristic of the MV⁺• radical developed. Molecular hydrogen was detected after 30 min, and over 60 turnovers of hydrogen were produced after 8 h of the photoirradiation. The activity of Pt1 in sensitizing photoinduced hydrogen production was examined by using [Co(dmgh)₂(py)Cl] as a catalyst. Photoirradiation of a degassed solution of Pt1 (2 × 10⁻⁵ M), Co³⁺ catalyst (2 × 10⁻⁴ M) and TEOA (0.1 M) with visible light was found to give approximately 500 turnovers of hydrogen after 6 h (light source: 300 W Oriel xenon lamp, λ > 400 nm).

In 2011, Che and co-workers reported photophysical and cytotoxicity studies of a series of [Pt(C∧N∧N)(NHC)]⁺ compounds (wherein NHC = N-heterocyclic carbene) (figure 9). The structurally simplest one was tested for its efficiency to sensitize light-induced hydrogen production. Our preliminary findings revealed that visible light irradiation of a degassed solution of [Pt(C∧N∧N)(NHC)]⁺ (2 × 10⁻⁵ M), Co³⁺ catalyst (2 × 10⁻⁴ M) and TEOA (0.1 M) gave about 420 turnovers of hydrogen after 4 h of irradiation (light source: 300 W Oriel xenon lamp, λ > 400 nm).

Recently, Che and co-workers reported a series of cyclometalated platinum(II) compounds for singlet oxygen generation [29]. A series of [Pt(Thpy)(PPh₃)X]⁺⁺ (HThpy = 2-(2'-thienyl)pyridine; X = Cl⁻, CH₃CN) and [Pt(Thpy)(HThpy)Y]⁺⁺ (Y = Cl⁻, pyridine) (figure 10) compounds were prepared. These compounds are emissive in degassed acetonitrile at room temperature with emission quantum yields and emission lifetimes up to 0.38 and 26 μs, respectively. The emission maxima are at 556 and 598 nm, which are assigned to excited states with mixed triplet metal-to-ligand charge transfer (³MLCT) and intraligand (³IL) characters. Upon exposure to air, there
is a substantial decrease in emission intensity and lifetime, indicating that the emissive triplet states of these platinum(II) compounds are effectively quenched by oxygen. By measuring the near-infrared luminescence signal intensity of $^{1}\text{O}_2$ at 1270 nm, the quantum yields of generating singlet oxygen by these compounds were estimated to be in the range of 0.42–0.95.

In another study, Che and co-workers reported a panel of cyclometalated platinum(II) compounds containing doubly deprotonated $\text{R}'\text{C}^\wedge\text{N}^\wedge\text{C}-\text{R}''$ ligands (2,6-diphenylpyridine derivatives) functionalized with carbazole, fluorene and thiophene units (figure 11) [30]. In non-degassed dimethylsulfoxide (DMSO) solutions, the emission intensity of these compounds, in each case, was gradually enhanced by approximately 10 times upon standing under ambient conditions. Such an enhancement of emission was only observed in DMSO but not in CH$_2$Cl$_2$, CHCl$_3$, acetone, CH$_3$CN, tetrahydrofuran (THF) or N,N-dimethylformamide. It should be noted that in the absence of light there was no such change in emission intensity. Gas chromatographic experiments revealed the formation of dimethyl sulfone. Thus, the emission enhancement can be explained by the consumption of dissolved oxygen which is needed for the generation of singlet oxygen. Subsequent oxidation of the solvent DMSO by the generated singlet oxygen greatly lowered the amount of dissolved oxygen in solution. As the emission quenching by dissolved oxygen was greatly reduced, the emission quantum yields and lifetimes of these compounds in the DMSO solution were enhanced.

Owing to the promising findings on singlet oxygen generation sensitized by the luminescent platinum(II) compounds, we examined their catalytic activities in light-induced aerobic functionalization of C–H bonds. We started the investigation by using [Pt($\text{C}^\wedge\text{N}^\wedge\text{N})(\text{NHC})]^{\text{+}}$ depicted in figure 9 as a photocatalyst. Upon visible light irradiation of a mixture of dibenzylamine and [Pt($\text{C}^\wedge\text{N}^\wedge\text{N})(\text{NHC})]^{\text{+}}$ (0.2 mol%) in CH$_3$CN under aerobic conditions, the secondary amine was found to be converted to imine with 123 turnovers in 1 h. As depicted in figure 12, three other amines were also effectively converted to give corresponding products with 87–142 turnovers by this protocol. We envision that photosensitization of singlet oxygen by platinum(II) compounds can be a useful tool for light-induced aerobic functionalization of a broad array of substrates.

3. Photoinduced catalytic properties of gold(III) compounds

The development of luminescent gold(III) compounds is at a much slower pace than that of the platinum(II) counterparts. An intrinsic reason is the presence of energetically low-lying gold(III) 5d$\text{x}^2$ – $\text{y}^2$ orbital which is anti-bonding with regard to gold–ligand $\sigma$ bond; hence, population of an electron in this orbital provides a facile non-radiative pathway for triplet excited state(s) to singlet ground state without emission. This intrinsic characteristic feature of gold(III) compounds could be an important reason accounting for the sparse gold(III) photochemistry reported in the literature.
Figure 12. Light-induced oxidative C–H bond functionalization reactions catalysed by \([\text{Pt}(\text{C}^\wedge\text{N}^\wedge\text{N})(\text{NHC})]^+\) (TON, turnover). Light source: 300 W Oriel xenon lamp, \(\lambda > 400\) nm.

Figure 13. Gold(III) compounds reported by Che and co-workers (Au1, Au2 and Au3). Au4 was reported both by Che and co-workers and by Yam and co-workers.

In order for gold(III) compounds to display strong and long-lived emission, the energy of the 5\(d_x^2 − y^2\) orbital has to be increased so that the potentially emissive IL and/or MLCT states would become low lying. As revealed by the literature reports on luminescent gold(III) compounds, the most efficient strategy is to use strong \(\sigma\)-donor ligands such as acetylide or carbene to increase the energy of the 5\(d_x^2 − y^2\) orbital. With the incorporation of acetylide [31,32] or N-heterocyclic carbene [33,34] ligands into gold(III) compounds supported by \(\text{C}^\wedge\text{N}^\wedge\text{C}\) or \(\text{C}^\wedge\text{N}\) type ligands, various organogold(III) compounds are emissive in solution with emission quantum yields up to 0.11.

One of the earliest works on gold(III) photochemistry was reported by Che and co-workers in 1994 [35]. This report described a luminescent gold(III) compound with a \(\text{C}^\wedge\text{N}^\wedge\text{N}\) tridentate ligand (Au1) as shown in figure 13. Compound Au1 is emissive in acetonitrile with emission maxima at 550 nm with emission quantum yield and lifetime of \(4 \times 10^{-4}\) and 0.55 \(\mu\)s, respectively. The emission of Au1 shows vibronic structures with spacings of approximately 1400 cm\(^{-1}\), attributed to the stretching frequencies of \(\text{C}=\text{C}\) and \(\text{C}=\text{N}\) bonds of the cyclometalated ligand. The presence of vibronically structured emission band and submicrosecond emission lifetime altogether suggest that the emission of Au1 originates from a triplet metal perturbed IL excited state. An important finding from this work is the high excited state reduction potential of Au1, being 2.2 V versus normal hydrogen electrode (NHE), indicating that Au1 is a strong photooxidant. The oxidizing power of Au1 in its triplet state had also been demonstrated by two photochemical reactions. Time-resolved absorption spectrum of a mixture of Au1 and 1,4-dimethoxybenzene in acetonitrile measured at 12 \(\mu\)s after excitation with a 355 nm laser pulse showed the formation of radical cation of 1,4-dimethoxybenzene. The strong oxidizing property of excited Au1 compound was also reflected by its ability to oxidatively cleave the C–H bonds of THF.

Another work revealing the rich photochemistry of organogold(III) compounds has recently been reported by Che and co-workers [34]. This work describes the photophysical and photochemical properties of two gold(III) compounds Au2 and Au3 (figure 13). With reference to the model compound Au4 reported by Yam and co-workers [33] and by Che and co-workers [36],
the luminescent properties of \([\text{Au}(\text{C}^\wedge\text{N}^\wedge\text{C})(\text{NHC})]^+\) can be improved by extending the \(\pi\)-conjugation of the phenyl ring. Compounds \(\text{Au}_2\) and \(\text{Au}_3\) are emissive in solution at room temperature with emission maxima at 519–521 nm. The emission quantum yields of \(\text{Au}_2\) and \(\text{Au}_3\) are 0.11 and 0.05, respectively, both of which are much higher than that of \(\text{Au}_4\) (0.004). What is more impressive is the enhancement in the emission lifetime. The triplet excited state lifetimes of \(\text{Au}_2\) and \(\text{Au}_3\) in degassed dichloromethane are 506 and 282 \(\mu\)s, respectively, which are much longer than that reported for \(\text{Au}_4\) (0.6 \(\mu\)s). The enhancement in excited state lifetime could be attributed to the rigidity of the extended \(\pi\)-conjugated cyclometalated \(\text{C}^\wedge\text{N}^\wedge\text{C}\) ligand, thereby greatly minimizing the structural distortion of the excited state. Notably, the emission of \(\text{Au}_2\) is completely quenched under aerobic condition. The quenching effect can be seen from figure 14, revealing the dramatic decrease in emission intensity upon exposing a degassed solution of \(\text{Au}_2\) to air. The high sensitivity of emission of \(\text{Au}_2\) towards oxygen renders \(\text{Au}_2\) to be a potentially good oxygen sensor. By combining electrochemical and emission data, the excited state reduction potential of \(\text{Au}_2\) was estimated to be 1.43 V versus NHE, suggesting that \(\text{Au}_2\) is a powerful photooxidant.

With such a long-lived excited state, \(\text{Au}_2\) was also found capable of catalysing light-induced oxidative \(\text{C}−\text{H}\) bond functionalization and hydrogen production reactions. In the presence of light irradiation, \(\text{Au}_2\) could effectively catalyse the aerobic oxidation of secondary amines to imines and \(\text{C}−\text{H}\) bond functionalization of tertiary amines with nucleophiles such as cyanide and nitromethane (figure 15). The results described in To et al. [34] could be favourably compared with the related literature findings when considering the low catalyst loading used (0.15 mol%). While luminescent platinum(II) compounds are well documented to sensitize light-induced hydrogen production from aqueous medium, there has been no report on using gold(III) compounds for light-induced hydrogen production prior to the report of To et al. [34]. Recently, \(\text{Au}_2\) was found to sensitize photoinduced hydrogen production by irradiating a degassed acetonitrile/water mixture of \(\text{Au}_2\) \((2 \times 10^{-5} \text{ M})\), \([\text{Co(dmgH)}_2\text{(py)}\text{Cl}]\) \((2 \times 10^{-4} \text{ M})\) and TEOA (0.1 M). The yellow colour of the reduction of cobalt(III) to cobalt(II) appeared within 30 s. Approximately 350 turnovers of hydrogen were generated after 4 h of irradiation. With the long-lived excited state and prominent photochemistry displayed by \(\text{Au}_2\), this report highlights the bright prospect of photophysics and photochemistry of organogold(III) compounds.

4. Photoinduced cytotoxic properties of platinum(II) compounds

Most of the reported phosphorescent platinum(II) compounds are highly emissive in visible spectral region, with long-lived triplet MLCT and/or IL charge transfer excited states, and high emission quantum yields. Because of their excellent luminescent properties, they have...
Figure 15. Photoinduced catalytic reactions of Au2. Light source: 300 W Oriel xenon lamp, $\lambda > 385$ nm.

Figure 16. Platinum(II) compounds examined by Che and co-workers for photoinduced cytotoxicity.

been used as phosphorescent dopants in organic light-emitting diodes [5,6]. The photophysical properties of cyclometalated platinum(II) compounds containing tridentate $C^\wedge N^\wedge N$ ligands (where $HC^\wedge N^\wedge N = 6$-phenyl-2,2$'$-bipyridine or its derivatives), or bidentate $C^\wedge N$ ligands (where $HC^\wedge N = 2$-phenylpyridine or its derivatives) have extensively been studied [8]. Their biological properties and applications as turn-on probes for (specific) proteins [37,38], enzyme poisons [39], G-quadruplex DNA stabilizers [40] and PDT agents [29] have recently been reported as well.

In a recent study, cyclometalated platinum(II) compounds containing thiophene moiety (figure 16, Pt3 and Pt4) were found to display high emission quantum yields (up to 0.38) and long emission lifetimes (24 $\mu$s) [29]. Time-resolved laser-induced emission measurements at the near-infrared region of 1270 nm revealed that the quantum yields for the generation of $1O_2$ in acetonitrile are 0.82 and 0.95 for Pt3 and Pt4, respectively.

Fluorescence microscopic experiments revealed that Pt4 could successfully enter into cancer cells and localize mainly in mitochondria (figure 17). We found that for cells treated with Pt4 irradiated with visible light (1 mW cm$^{-2}$) for 30 min and further incubated for 47.5 h in the dark, the cytotoxic IC$_{50}$ values (concentration of inhibiting 50 per cent cellular growth compared with control) of Pt3 and Pt4 were 3.29 and 1.78 $\mu$M, respectively. As a comparison, the corresponding IC$_{50}$ values without light irradiation were 21.1 and 6.12 $\mu$M (figure 18).
Figure 17. Live cell images of HeLa cells incubated with Pt3 at 5 μM for 1 h showing (a) fluorescence emitted by Pt4 (excitation wavelength = 470 nm); (b) nucleus stained with Hoechst 33342 (upper, excitation wavelength = 365 nm) or mitochondria stained with MitoTracker Red (lower, excitation wavelength = 546 nm). (c) Overlay of fluorescence images of (a) and (b) [29]. (Online version in colour.)

Figure 18. Plot of percentage viability of HeLa cells as a function of concentration of Pt4 in the dark (black line) and after irradiation with visible light (grey (red) line). Light source: fluorescent lamp, 1 mW cm⁻² [29]. (Online version in colour.)

Platinum(II) compounds containing tridentate cyclometalated and N-heterocyclic carbene ligands (figure 19) are stable in physiological conditions and are strongly emissive in organic solvents. Contrary to the classical platinum(II) anti-cancer drugs such as cisplatin and oxaliplatin, Pt5 does not accumulate in the nucleus but preferentially accumulates in cytoplasmic structures including sites where active survivin, an inhibitor of apoptosis, is located (figure 20) [41].
Figure 19. Platinum(II) compounds containing tridentate cyclometalated and N-heterocyclic carbene ligands.

Figure 20. Fluorescent microscopic examination of Pt5 in the same batch of HeLa cells either co-incubated with MitoTracker (top, excitation wavelength = 546 nm), Hoechst 33342 (middle, excitation wavelength = 365 nm) or LysoTracker (bottom, excitation wavelength = 546 nm), showing that the majority of Pt5 can be co-localized with MitoTracker [29]. (Online version in colour.)

The platinum compounds Pt5–Pt10 are highly cytotoxic (in the dark) and display a high specificity to cancerous cells (table 1). Compound Pt5 was found to display in vitro anti-cancer activity towards three cancer cell lines with IC50 values of 0.057–0.77 μM. Notably, this compound is less cytotoxic to the normal lung fibroblast cell line (CCD-19Lu) with an IC50 value of 11.6 μM, the latter is 232-fold higher than that towards HeLa cells.
Moreover, compound \( \text{Au5} \) than the normal lung fibroblast cell line (CCD-19Lu) (table 2). In addition, upon exposure to visible light for only 1 h, the cytotoxic IC\(_{50} \) value (24 h of incubation) of 

\[
\text{C2,6-diphenylpyridine) \[41–43\].}
\]

could effectively poison topoisomerase I and suppress tumour growth in nude mice models \[36\]. The cytotoxic IC\(_{50} \) value of 

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\text{change from 7.7 to 3.4 } \mu\text{M} \text{ (figure 21). The cytotoxic IC50 value of Au5} \text{ compounds against selected human cell lines.}
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As \( \text{Pt5} \) is strongly phosphorescent, its phototoxicity towards cancer cells was also examined (figure 21). The cytotoxic IC\(_{50} \) value of \( \text{Pt5} \) in the presence of visible light was found to be 0.63 \( \mu\text{M} \), which is about 6.5-fold more cytotoxic than that in the dark conditions (IC\(_{50} = 4.1 \mu\text{M}\)).

5. Photoinduced cytotoxic properties of cyclometalated gold(III) compounds

The use of photosensitizers having a high solution stability is a prerequisite for PDT in general. Yet, various gold(III) compounds are usually unstable under physiological conditions due to the ease of reduction of the metal centre as well as the high reactivity towards thiols (like glutathione). All of these properties have hampered the development of gold(III) compounds for therapeutic usages. We previously employed a series of dianionic tridentate and tetradeutate ligands to stabilize gold(III) ion and prepared various kinds of physiologically stable gold(III) compounds \[42\]. Some of these compounds were found to display promising in vitro and in vivo anti-cancer activities. The two representative classes of gold(III) compounds are the ones with porphyrin ligands and those with doubly deprotonated C\(^{\pi}\)N\(^{\pi}\)C ligands (where HC\(^{\pi}\)N\(^{\pi}\)CH = 2,6-diphenylpyridine) \[41–43\].

In a recent study, we reported that various [Au(C\(^{\pi}\)N\(^{\pi}\)C)(NHC)]\(^+\) compounds (figure 22) could effectively poison topoisomerase I and suppress tumour growth in nude mice models \[36\]. Moreover, compound \( \text{Au5} \) displays a higher selectivity (167-fold) towards tumour cell line (NCI-H460) than the normal lung fibroblast cell line (CCD-19Lu) (table 2). In addition, upon exposure to visible light for only 1 h, the cytotoxic IC\(_{50} \) value (24 h of incubation) of \( \text{Au5} \) was found to change from 7.7 to 3.4 \( \mu\text{M} \).

As discussed in §3, the [Au(C\(^{\pi}\)N\(^{\pi}\)C)(NHC)]\(^+\) compounds containing extended \( \pi \)-conjugated C\(^{\pi}\)N\(^{\pi}\)C ligands (\( \text{Au2} \) and \( \text{Au3} \)) have long phosphorescence lifetimes (506 \( \mu\text{s} \) for \( \text{Au2} \); 282 \( \mu\text{s} \) for \( \text{Au3} \)) and high emission quantum yields (11.4% for \( \text{Au2} \); 5.5% for \( \text{Au3} \)) \[34\], making them good candidates as effective photooxidative catalysts and photosensitizers for PDT. Although
Figure 21. The cytotoxicity of Pt5 in the presence or absence of light. Light source: fluorescent lamp, 1 mW cm$^{-2}$. (Online version in colour.)

Figure 22. Gold(III) compounds with N-heterocyclic carbene ligands (Au5–Au9).

Figure 23. When nitrogen was blown over living cells incubated with Au3 in buffer solution (a), cancer cells became yellowish green (b) illuminated under a fluorescent microscope. Excitation wavelength = 368 nm [34]. (Online version in colour.)
Figure 24. Cytotoxicity profiles (24 h) of (a) Au$_3$ and (b) Au$_2$ in the presence or absence of light. Light source: fluorescent lamp, 1 mW cm$^{-2}$. (Online version in colour.)

Figure 25. Cytotoxicity profiles (24 h) of Au$_9$ in the presence or absence of light. Light source: fluorescent lamp, 1 mW cm$^{-2}$. (Online version in colour.)
these compounds are non-emissive at room temperature due to efficient quenching of their phosphorescence by oxygen, they could be used as luminescent probes for cell imaging under hypoxia conditions. Yellow–green colour image of the cells coming from the phosphorescence of \textbf{Au3} developed in hypoxia conditions was recorded (figure 23). This result reveals that \textbf{Au3} could effectively enter into cancer cells upon incubation.

Combining the advantages of the gold(III) compounds in the generation of singlet oxygen and their capability of entering into cancer cells, the phototoxicity of \textbf{Au2} and \textbf{Au3} has been examined. After exposure to visible light for 1 h, different cytotoxic profiles (24 h) of \textbf{Au2} and \textbf{Au3} were observed compared to those in the absence of light. The cytotoxic IC$_{50}$ values for \textbf{Au2} and \textbf{Au3} in the presence of light are 0.10 and 0.13 $\mu$M, respectively. Both of these values are about 10- to 12-fold smaller than the corresponding IC$_{50}$ values of 1.1 and 1.7 $\mu$M, respectively, in the absence of light (figure 24).

We have modified the extended $\pi$-conjugated C\^\text{\textdegree}N\^C ligand system by incorporation of a phenyl ring on the pyridine group (\textbf{Au9}, figure 22) and tested for the phototoxicity of the resulting gold(III) compound. The IC$_{50}$ value of this gold(III) compound was found to be 1.6 $\mu$M in the presence of visible light (figure 25). Compound \textbf{Au9} in dark conditions is also cytotoxic (IC$_{50}$ value = 2.6 $\mu$M), consistent with the previous results that the cytotoxicity could be maintained even if the C\^\text{\textdegree}N\^C ligand is modified (e.g. \textbf{Au2} and \textbf{Au3}). These exciting results reveal the bright clinical prospects of combining cytotoxicity and phototoxicity of [Au(C\^\text{\textdegree}N\^C)(NHC)]$^+$ compounds.

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