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Faculty of Science

Ph.D. Thesis in Inorganic Chemistry

Synthesis and Characterization of Potentially Biological Active Cyclometallated Organoplatinum(II) Complexes

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November 2011

In the Name of God

DECLARATION

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تقدیم به همسر مهربانم

کوثر بی مانند زندگیم و بهترین هدیه الهی ام

او که همچون شمعی سوخت تا روشنی هستی ام باشد.

سپاس

سپاس بی نهایت خداوند مهربان که در همه مراحل زندگی پشتیبان و راهنمای بی منت برایم بوده و هست. بر خود لازم می دانم که از خانواده ام و خانواده همسرم که صمیمانه و خالصانه مسیر پیشرفت مرا هموار کردند تشکر کنم. سپاس ویژه از همسر مهربان و فداکارم که در همه مراحل علمی، یار و همراه من بوده و هست و به پاس قدردانی از همه این پایان نامه به ایشان تقدیم می دارم.

بر خود لازم می دانم از آقای دکتر رشیدی که با صبر و ممارست در طی ۸ سال تلاش زیادی در جهت باروری علمی و اخلاقی من نموده تشکر ویژه ای داشته باشم.

سپاس ویژه از جناب آقای دکتر اسماعیل بیگ بواسطه پشتیبانی ها و حمایتهای همه جانبه که درس صبوری و اخلاق را از ایشان آموختم.

سپاس ویژه از همه اساتیدی که برای رسیدن من به این مرحله تلاش و کوشش نمودند و چون شمع نورانی در راستای ترقی من نور فشانی کردند تا راهگشای تاریکیهای زندگیم باشند. امیدوارم شاگرد و فرزند خوبی برای همه اساتیدم باقی بمانم.

بر خود لازم می دانم از آقای پروفسور کپلر و گالانسکی از دانشگاه وین با حمایتهای مالی و تکنیکی در هر چه پر بار نمودن این پایان نامه سهم بسزایی داشتند و بدون هیچ انتظاری از هیچگونه لطفی فرو گزار نکردند قدردانی ویژه نمایم.

از همه دوستان عزیزی که در جهت پیشرفت من تلاش نمودند قدردانی می نمایم به ویژه آقای محسن گلبن حقیقی که اشتیاق ویژه ایشان در خودسازی و آموزش و پژوهش برایم الگو بوده است.

در پایان لازم است ذکر نمایم که شیرین ترین و فراموش نشدنی خاطرات من در دانشگاه شیراز و مردم خوب و مهربان شیراز است. امیدوارم روزی بتوانم لطف همه کسانی که به گردن من حقی دارند جبران نمایم.

حمید رضا سموئی

ABSTRACT

Synthesis and Characterization of Potentially Biological Active Cyclometallated Organoplatinum(II) Complexes

By

Hamidreza Samouei

This work is presented in five parts. In the first part preparation of the starting complex [Pt(C^N)Cl(dmso)], **1**, in which C^N = N(1),C(2′)-chelated, deprotonated 2-phenylpyridine, and dmso = dimethylsulfoxide, and its reaction with 1 equiv of the biphosphine ligands bis(diphenylphosphino)amine, dppa, or bis(diphenylphosphino)methane, dppm, to give the complex [Pt(C^N)Cl(dppa)], **2**, or [Pt(C^N)Cl(dppm)], **3**, respectively are described. Careful 1- and 2D-NMR and conductivity measurements confirm that the structure of complexes **2** and **3** in solution is unusual neutral penta-coordinated. However, X-ray crystallography indicated that the solid state structure of each of the complexes **2** and **3** is comprised of a cationic platinum(II) species having a common square-planar geometry with a Cl^- counter-anion. Penta-coordinated molecules of complex **2** in solution also form a rare type of N-H···Pt intermolecular hydrogen bonding. Complexes [Pt(C^N)(dppa)](PF_6), **4**, and [Pt(C^N)(dppm)](PF_6), **5**, prepared by the reaction of complexes **2** and **3** with NH_4PF_6, having PF_6^- counter-anion with no coordinating ability. These complexes were found to be ionic in solution and the N-H···Pt intermolecular interaction in solution of complex **2** was vanished in

the solution of complex **4**. Furthermore, the pharmacological effects of complexes **2** and **3** were evaluated in terms of their proteasome-inhibitory and apoptosis-inducing activities under *in vitro* and *in vivo* conditions. both complexes **2** and **3** showed significant proteasome-inhibitory activity against purified 20S proteasome, while complex **3** demonstrated superior inhibitory activity against cellular 26S proteasome. Luminescent properties of both complexes **2** and **3** and their binding interaction with herring sperm DNA has been investigated by fluorimeteric emission study using ethidium bromide (EB) as a fluoresce probe. Our results strongly suggest that the Pt^{II}-containing biphosphine complexes that target the tumor proteasome have potential to be further investigated as potential anticancer drugs.

In part two the starting complex **1** was reacted with either 1 or 0.5 equiv of 1,1'-bis(diphenylphosphino)ferrocene, dppf, to give the cyclometalated diplatinum(II) complex $[Pt_2(C\hat{}N)_2Cl_2(\mu\text{-dppf})]$, **7**. Complex **7** was fully characterized in detail in solution by using multinuclear NMR spectroscopy (^{1}H, ^{13}C, ^{31}P, and ^{195}Pt) supported by a number of 2D NMR experiments, while the structure in solid state was determined by X-ray crystallography. Cytotoxicity of the complex **7** was studied in three resisted human cancer cell lines derived from ovarian carcinoma(CH1), lung carcinoma(A549), and colon carcinoma (SW480) by means of the MTT assay (MTT = 3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyl-2H-tetrazolium bromide).

Part three describes reaction of complex **1** with 1 equiv of either 2-(diphenylphosphinoamino)pyridine , PPh_2NHPy, or 2-diphenylphosphino pyridine, PPh_2Py, giving the complex $[Pt(C\hat{}N)(PPh_2NHPy)]Cl$, **8**, or $[Pt(C\hat{}N)Cl(PPh_2Py\text{-}\kappa^1P)]$, **9**, respectively. Careful multinuclear 1D NMR spectroscopy (^{1}H, ^{13}C, ^{31}P, and ^{195}Pt), 2D NMR and conductivity measurements show structure for complex **8** in solution is neutral penta-coordinated, while complex **9** is found to be neutral four coordinate. X-ray crystallography indicated that the solid state structure of complex **8** is comprised of a cationic platinum(II) species having a common square-planar geometry with a Cl^- counter-anion. The complex **8** forms a rare type of intermolecular N-H···Pt hydrogen bonding in

solution. Cytotoxicity properties of the complexes **8** and **9** were studied in three resisted human cancer cell lines.

In part four, the reaction of complex **1** with 1 equiv of either 1,3,5-triaza-7-phosphaadamantane, PTA, or triphenyl phosphine, PPh_3, to give the complex [Pt(C^N)Cl(PTA)], **10**, or [Pt(C^N)Cl(PPh_3)], **11**, respectively, is described. Multinuclear 1 and 2D NMR spectroscopy show the structures of complexes **10** or **11** in solution being neutral four coordinate. The X-ray crystallography indicated that the solid-state structure of complex **10** is comprised of a common square-planar geometry around platinum(II). Cytotoxicity of the complexes **10** and **11** were studied in three human cancer cell lines.

Part five describes the reaction of complex $PtCl(dmso)_2$, with NaI to form the starting complex *cis/trans* [$Pt_2I_4(dmso)_2$], **12**. Complex **12** was consequently reacted with 2 equiv of 2-(diphenylphosphinoamino)pyridine, PPh_2NHPy, P^N, to form complex [PtI_2(P^N)], **13**. Complex **13** was fully characterized by 1- and 2D multinuclear NMR, ESI mass and elemental analysis. Complex **13** is an unprecedented example of a platinum(II) complex with simultaneous formation of intermolecular NH···I-Pt and CH···I-Pt H-bondings (with neighboring platinum center) and an intramolecular CH···Pt hydrogen bonding in solid state. There are indications showing that the complex in solution probably forms different kinds of H-bonding interactions.

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List of Abbreviations:

C^N = deprotonated 2-phenylpyridine

COSY = Correlation Spectroscopy

Cp = cyclopentadienyl

DEPT = Distortion-less Enhancement by Polarization Transfer

dmso = dimethylsulfoxide

dppa = bis(diphenylphosphino)amine

dppf = 1,1′-bis(diphenylphosphino)ferrocene

dppm = bis(diphenylphosphino)methane

DQF-COSY = Double Quantum Filtered Correlation Spectroscopy

EB = ethidium bromide

ESI-Mass = electrospray ionization mass spectrometry

H&E = Hematoxylin and eosin

HC^N = 2-phenylpyridine

HETCOR = Heteronuclear Correlation

HMBC = Heteronuclear Multiple Bond Correlation

HMQC = Heteronuclear Multiple Quantum Correlation

HSQC = Heteronuclear Single Quantum Correlation

IC_{50} = 50% inhibitory concentrations

MTT = 3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyl-2H-tetrazolium bromide

PTA = 1,3,4-triaza-7-phosphatricyclo[3.3.1.1]decane

RT = room temperature

TUNEL = Terminal deoxyribonucleotidyl transferase-mediated dUTP nick end-labeling

Chapter One

Chapter One

Introduction

1.1. General Remarks

Organometallic compounds are those compounds having bonds between one or more metal atoms and one or more carbon atoms of an organyl group. They are classified by prefixing the metal with *organo*-, *e.g.* organoplatinum compounds. In addition to the traditional metals and semimetals, elements such as boron, silicon, arsenic and selenium are considered to form organometallic compounds.

Depending mostly on the nature of metallic ion and somewhat on the nature of the organic compound, the character of the bond may either be ionic or covalent. Organometallic compounds with bonds that have characters in between ionic and covalent are very important in industry and medicine, as they are both relatively stable in solutions and relatively ionic to undergo reactions.

1.2. Organoplatinum Complexes

The first compound containing an unsaturated hydrocarbon attached to a metal, and indeed the first organometallic compound, if one excludes the cyanides, was $[Pt(C_2H_4)Cl_2]_2$, discovered by the Danish chemist W. C. Zeise as long ago as 1827 and followed 4 years later by the salt which bears his name, $K[Pt(C_2H_4)Cl_3].H_2O$.

Ever since that time, platinum has been an important element in organometallic chemistry because it forms a wide range of organometallic compounds that are

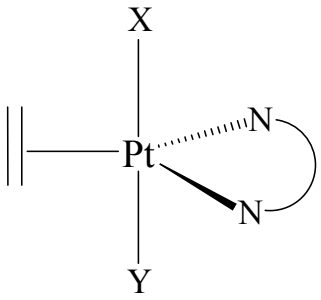
kinetically sufficiently inert to enable them to be isolated and characterized. The development of NMR for platinum, that 33.7% of which is present in nature as the 195 isotope which has a nuclear spin of ½ , has been attractive because of the possibility of observing coupling between the metal and other nuclei. The presence, or absence, of such coupling provides valuable evidence on which to base structural conclusions as well as to make mechanistic suggestions for the reactions of organoplatinum complexes.

1.3. Geometry in Platinum Complexes

The common geometries for platinum complexes are four and six coordinations. However, study on the uncommon five-coordinated systems is desirable, because of their role as intermediate in many mechanistic investigations concerning reactions proceeding by associative, involving four-coordinated complexes, or dissociative, involving six-coordinated complexes. The geometrical studies on trigonal bipyramid and square pyramid structures as two extreme possible structures are also of great interest. As well recent reports show higher antitumor activity of five-coordinated systems than that of four-coordinated platinum complexes.[1-3] Usually the five-coordinated systems are not stable enough and are difficult to be separated and special electronic and structural conditions are required in order to stabilize the related complexes.[4,5]

Five-coordinate trigonal bipyramydal Pt^{II} complexes [Pt(N^N)(olefin)XY] (X = halide, Y = halide or hydrocarbyl group; N^N = bidentate nitrogen ligand) represent a well-characterized class of platinum compounds (Scheme 1.1).[6,7] The presence on the trigonal plane of a π-acceptor olefin ligand which draws electron charge from the metal center, so favoring the coordination of both ends of the bidentate nitrogen donor, plays a crucial role in stabilizing the five-coordinate geometry in these species. When N^N = 2,9-Me_2-1,10-phenanthroline the five-coordinate complexes are particularly stable if compared to analogous species with other bidentate nitrogen ligands. Such a stabilization stems from the release, in the five-coordinate species, of the steric interaction between the ortho methyl

substituents of the phenanthroline and the *cis*-halogen ligands built up in square planar $[Pt^{II}X_2(2,9\text{-}Me_2phen)]$[8] and octahedral $[Pt^{IV}X_4(2,9\text{-}Me_2\text{-}phen)]$[9] complexes.



Scheme 1.1.

Due to the steric strain, the $[PtX_2(2,9\text{-}Me_2\text{-}phen)]$ complexes easily dissociate one end of the phenanthroline chelate to form a T-shaped, coordinatively unsaturated, platinum species which are highly susceptible to addition reactions.[8,10] The addition product, $[Pt^{II}X_2(2,9\text{-}Me_2\text{-}phen)(L)]$, can be either a square-planar species, L = CO, PPh_3, Me_2SO, Me_2S, PhNO, Py, $NH_2(CH_2)_2CH_3$], with the phenanthroline ligand monocoordinate to the platinum center (with exchanging in solution of the donor nitrogen atoms at the coordination site) or, in the case of L = alkene[11] and alkyne,[12] a five-coordinate trigonal bipyramidal species with the halogen ions in the axial positions and the phenanthroline ligand acting as chelate and lying, together with the L ligand, in the trigonal plane.

Octahedral $[PtX_4(2,9\text{-}Me_2\text{-}phen)]$ complexes also react with excess C_2H_4 leading to reductive elimination of halogen (which, in turn, reacts with excess C_2H_4 to give 1,2-dihaloethane) and formation of the five-coordinate Pt^{II} species $[PtX_2(C_2H_4)(2,9\text{-}Me_2\text{-}phen)]$.

The formation of L_3PtX_2 is enthalpy favored and entropy disfavored. The relative thermodynamic stability of the L_3PtX_2 complexes is a function of ligand steric bulk; the smaller ligand gives the greater stability. The stereochemical rigidity of the L_3PtX_2 complexes is inversely proportional to ligand steric bulk: the larger ligand, the more rigid L_3PtX_2 complex.[13]

NMR spectroscopy alone cannot unambiguously determine the geometry of L_3PtX_2 species in solution, since the same A_2X pattern is expected for each of the four configurations (Figure 1.1). The solution geometries of the complexes are most likely distorted between square pyramidal and trigonal bipyramidal, consistent with the limited amount of structural data for L_3PtX_2 complexes.[13]

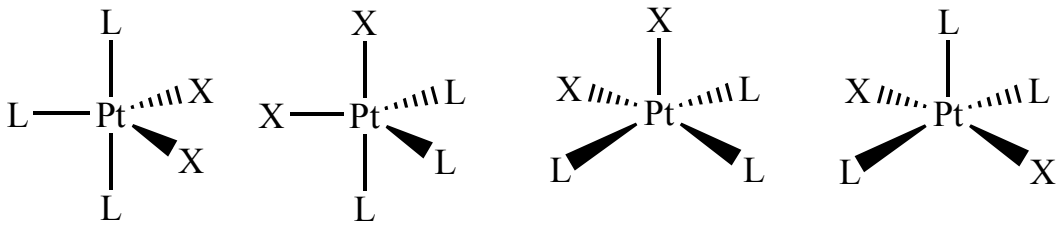


Figure 1.1. Possible structures for L_3PtX_2 complexes.

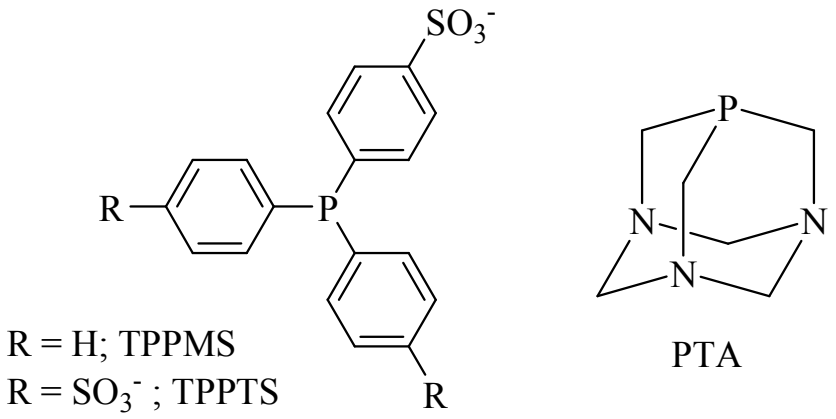
Detailed comparisons of these structures together with the solution NMR data for the L, Pt, X, complexes suggest that steric rather than electronic factors are dominant in determining the thermodynamic stability of the penta-coordinate complexes.[14]

1.4. Phosphine Ligands

Organophosphines are among the most common ancillary ligands used in organometallic chemistry, owing to their ability to stabilize low metal oxidation states and to the capacity to influence both steric and electronic properties of the catalytic species. One of the advantages of using phosphines as ligands is that they can be easily modified by changing the organic substituents, thus allowing the fine tuning of the electronic and steric properties of the metal complexes. In homogeneous catalysis, this can be a very useful tool in order to change the activity or selectivity of the catalyst. These advantages can be maintained and solubility in water can be added by modifying the phosphine structure by introducing polar substituents such as hydroxyl or amino functionalities or ionic

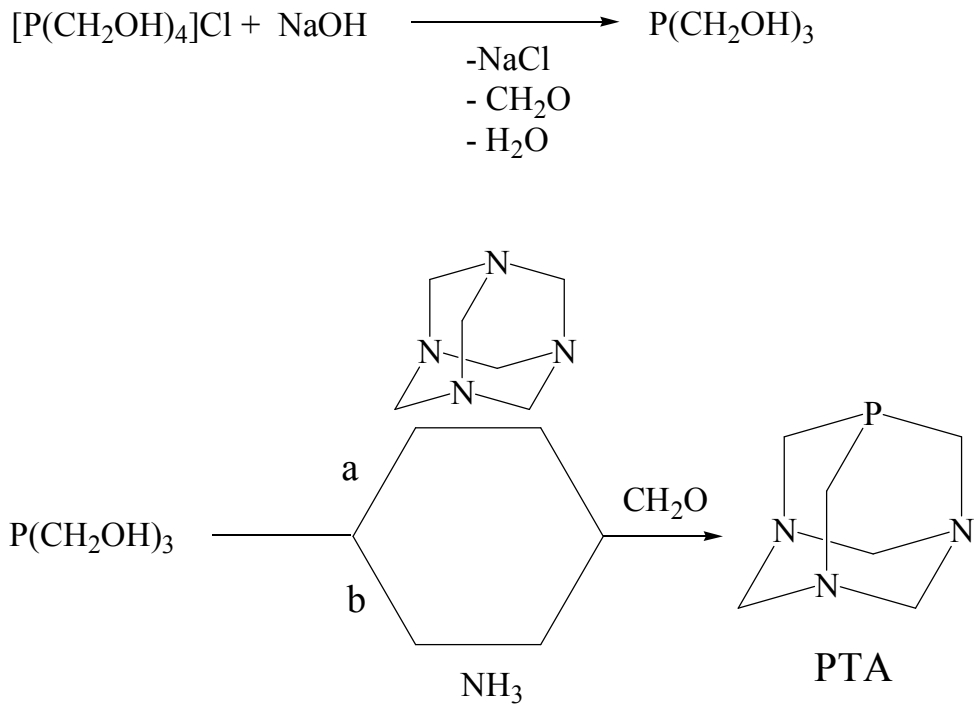
groups such as sulfonate, carboxylate and ammonium, just to mention some of the more important.[15]

Examples of water-soluble monodentate aryl phosphines are the sulfonated analogues of PPh_3, namely the monosulfonated TPPMS and tris-sulfonated TPPTS (Scheme 1.2) (for examples of sulfonated phosphines and applications of these ligands see among the others).[16-27] However, also bidentate diphosphines and tridentate tripodal phosphines are represented by their sulfonated derivatives.[17,18,26,27] A small number of water-soluble cage-like phosphines have been characterized including the well known Verkade-type bases used as ligands in a number of organic reactions.[28] Another cage adamantane like phosphine, 1,3,5-triaza-7-phosphaadamantane (Scheme 1.2) (usually abbreviated as PTA)[16-27] first reported in 1974 by Daigle et al. has been only sparingly used since its discovery.[29] Originally, the study aimed at synthesizing PTA was intended to create flame-proof polymers. However, with the recent search for water-soluble catalysts,[30-32] PTA and its derivatives have received renewed interest and the increasing number of coordination compounds containing this unique ligand and their applications in homogeneous aqueous biphasic catalysis makes the chemistry of PTA timely and worth to be reviewed.[15,33] In addition to use as ligand for biphasic catalysis, PTA and related species are important hydrophilic co-ligands in biological active transition metal compounds and are excellent ligands for preparing luminescent gold complexes.



Scheme 1.2.

The synthesis of PTA is a straightforward reaction involving the condensation of trishydroxymethylphosphine with formaldehyde and hexamethylenetetraamine in ice-water, the final yield being around 40% (Scheme 1.3, route (a)).[29] Alternatively, a solution of ammonia and formaldehyde can be used in place of hexamethylenetetraamine (Scheme 1.3, route (b)).[29] Later research showed that higher yielding preparations (65–80%) are obtained by initially forming trishydroxymethylphosphine $P(CH_2OH)_3$ in situ from reacting the commercially available and less expensive tetrakis(hydroxymethyl)phosphonium chloride $[P(CH_2OH)_4]Cl$ with sodium hydroxide.[34] PTA is generally isolated as an analytically pure compound when recrystallized from hot ethanol or acetone. The synthesis of PTA does not require an inert atmosphere as it is neither air or moisture sensitive. Furthermore, PTA is thermally stable, decomposing at temperatures higher than 260 °C. PTA is soluble in water (ca. 235 g/L), MeOH and EtOH, but less soluble in heavier alcohols such as 2-propanol or *n*-butanol and THF at room temperature.[34] The solubility of PTA in acidic media, following the formation of its protonated form, is even larger reaching approximately 350 g/L in 0.1 M HCl. Furthermore, PTA is soluble in DMSO, acetone, chloroform, dichloromethane, but not in hydrocarbons such as toluene, benzene or hexane.



Scheme 1.3.