Diruthenium Complexes for Bio-imaging

Group 11-11

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Abstract

Bio-imaging is an important part of cancer diagnosis as it allows for tumours to be viewed and located non-invasively through different methods. However all these methods come with their own problems, such as being incompatible with certain patients, or being harmful in large doses. As such, there is a need for a better method for bio-imaging, in the form of diruthenium complexes, which have certain properties which make it a promising candidate for use in bio-imaging. We have synthesised two diruthenium complexes, a diruthenium sawhorse complex and a diruthenium cationic complex, incorporating carboxylated chalcone into the molecules. This was done through a series of chemical reactions using different chemicals. Infrared spectroscopy and mass spectrometry confirmed the identity of our products. We had successfully synthesised both diruthenium complexes. However, further research must be conducted to ensure that the products are able to carry out its function in the human body without any implications.

Introduction

Bio-imaging is a technique commonly used in hospitals and clinics around the world to allow medical personnel to view biological processes in patients' bodies with 2- or 3-dimensional diagrams obtained through non-invasive diagnostic machines. The term 'non-invasive' refers to procedures that do not break the skin and do not come into contact with internal body cavities beyond that of natural orifices. Since bio-imaging's emergence in the 20th century as a way for doctors to inspect their patients for internal signs and symptoms of injuries, it developed and evolved many times and culminated in some frequently used and effective methods of scanning, namely Computerised Tomography (CT) and X-ray scans, Magnetic Resonance Imaging (MRI) and ultrasound scans.

Despite the success of the aforementioned methods, there are still some groups of patients excluded from the scope of such devices and scans due to various reasons. CT scans and X-rays used to be conducted by doctors for nearly every patient that they encountered, but in the early 20th century just a few years after X-ray was discovered in 1895 reports started surfacing of distressing side effects of long term exposure to such scans, forcing doctors and medical organisations to restrict and implement guidelines on the usage of X-rays (Fritzsche, Dong & Moeendarbary, 2018). Today, the limited number of scans a patient can undergo restricts the quality of diagnosis that doctors can give their patients. MRI are another feasible method of bio-imaging, but due to the nature of the scans, which utilise strong magnetic fields to image the body, patients with pacemakers, metallic prosthetics and implants cannot undergo the imaging process due to safety concerns when metallic objects enter the magnetic field generated (Brown, Cheng, Haacke, Thompson & Venkatesan, 2014). Ultrasound is one more but less common method of bio-imaging as it provides quite a low image quality in its scans and so are less preferred by medical professionals (Dhawan, 2003).

Looking at the problems bio-imaging is facing nowadays, we aim to utilise diruthenium complexes for bio-imaging to identify tumours and the like in patients' bodies. The imaging method we propose to be used is thermography, a form of bio-imaging that uses infrared rays to visualise a patient's body. Thermal imaging allows for multiple disease patterns to be imaged based on the body's temperature distribution, as humans radiate heat in the form of infrared rays (*Thermography in Medicine*). When we introduce our diruthenium complexes into the patient's body by in the form of a solution that he or she consumes, they will seek out and attach themselves to any probable tumours or abnormalities in the body, due to their unique chemosensing properties. They can then be identified on the thermographic scan as their heat signature will be different from that of the surrounding parts of the body, helping doctors to locate the possible tumour or abnormal growth (Cardone & Merla, 2017).

Diruthenium complexes are potential substances that can be used in the growing field of bio-imaging. They will be composed of ruthenium, chalcone, and carbonyl groups. The properties of the compounds that make up the molecule gives it unique properties which can be utilised for bio-imaging. Ruthenium in the complex will act as the core of the molecule which will hold the entire molecule together. It can also act as a probe for certain biological substances associated with tumour growth and the complex can be viewed using thermal imaging due to the high photothermal stability of ruthenium complexes (Lin, Zhao, Bo, Hao & Wang, 2018). In addition, research by Lin et al. (2018) revealed that diruthenium complexes are low in toxicity and can potentially be safe for humans. Chalcone also has target identification properties for certain biological molecules related to cancer, such as epidermal growth factor receptor, aurora kinase, and anaplastic-lymphoma kinase (Zhuang et al., 2017). The carbonyl groups allows the complex to be tracked by infrared rays due to its intense stretching vibration peak .

Objectives

To synthesise and characterise a novel diruthenium sawhorse complex and diruthenium cationic complex incorporating carboxylated chalcone

Apparatus and Materials

Apparatus	Chemicals
100mL conical flask 50mL measuring cylinder Electronic measuring scale Fume hood Magnetic Stirrer and Bar Suction filtration vacuum pump Whatman filter paper Filter funnel Schlenk tube	Sodium Hydroxide (NaOH) Deionised water 90% Ethanol 4-formylbenzoic acid ($C_8H_6O_3$) Acetophenone (C_8H_8O) 1M hydrochloric acid (HCl) Triruthenium dodecacarbonyl (Ru_3CO_{12}) Pyridine (C_5H_5N)

Hotplate with temperature probe Liebig condenser Recirculating chiller Round bottom flasks Rotary evaporator Heating mantle Fourier Transform Infrared Spectroscope Electrospray Ionization Mass Spectrometer	2,2'-bipyridine ($C_{10}H_8N_2$) Sodium tetraphenylborate ($NaBPh_4$)

Methods & Procedures

Synthesis of carboxylated chalcone ($C_{15}H_{11}OCOOH$)

1. 0.400g of solid sodium hydroxide (NaOH) was added to 25mL of water and 10mL of 90% ethanol solvent in a 100mL conical flask.
2. 0.60g of 4-formylbenzoic acid ($C_8H_6O_3$) was added to the reaction mixture and stirred at ambient temperature for 15 minutes.
3. 0.466mL of acetophenone (C_8H_8O) was added and the reaction mixture was further stirred for another 24 hours.
4. Reaction mixture was acidified with 10.0mL 1M HCl.
5. Carboxylated chalcone was retrieved by suction filtration and air-dried for another 24 hours

Synthesis of diruthenium sawhorse complexes

1. 100mg of triruthenium dodecacarbonyl (Ru_3CO_{12}) and 107mg of carboxylated chalcone ($C_{15}H_{11}OCOOH$) was added to 40mL of ethanol in a Schlenk tube.
2. The mixture was refluxed overnight for 18 hours.
3. The mixture was cooled down and 37.1µL of pyridine (C_5H_5N) was added to the reaction mixture and the reaction mixture was further stirred for 2 hours.
4. Diruthenium sawhorse complex was collected by evaporation of ethanol solvent and was air dried for a further 24 hours.

Synthesis of diruthenium cationic complexes

1. 340mg of diruthenium sawhorse complex and 109mg of 2,2'-bipyridine was added to 50mL of ethanol in a round bottom flask.
2. The mixture was stirred at 50 °C using a heating mantle overnight.
3. 120mg of sodium tetraphenylborate ($NaBPh_4$) was added to the reaction mixture.
4. Diruthenium cationic complex was collected by filtration and left to dry overnight.

Results and Discussion

Synthesis of Sawhorse complex

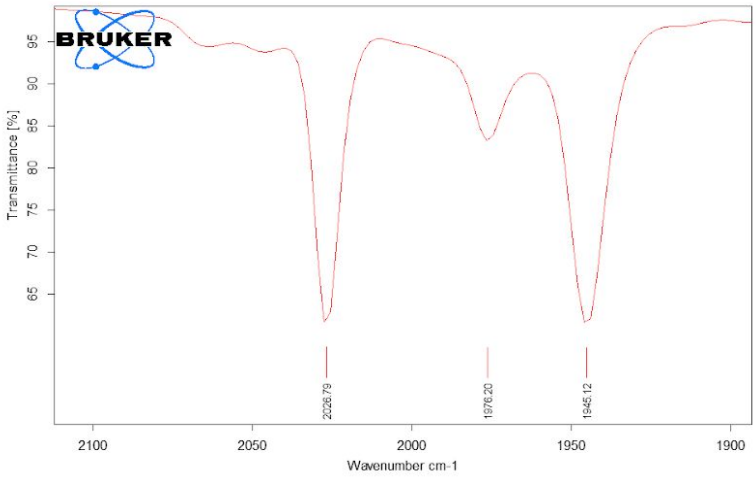


Figure 1: FTIR wave pattern of sawhorse

By Fourier Transformed Infrared (FTIR) Spectroscopy, we obtained the infrared (IR) graph (Figure 1) where three distinct peaks can be observed at wavenumbers 2027cm^{-1}, 1976cm^{-1} and 1945 cm^{-1} respectively. The strong-weak-strong pattern as well as the wavenumbers observed, is closely associated with the sawhorse structure in other organometallic compounds with ruthenium as the transition metal (Jiang et al., 2017).

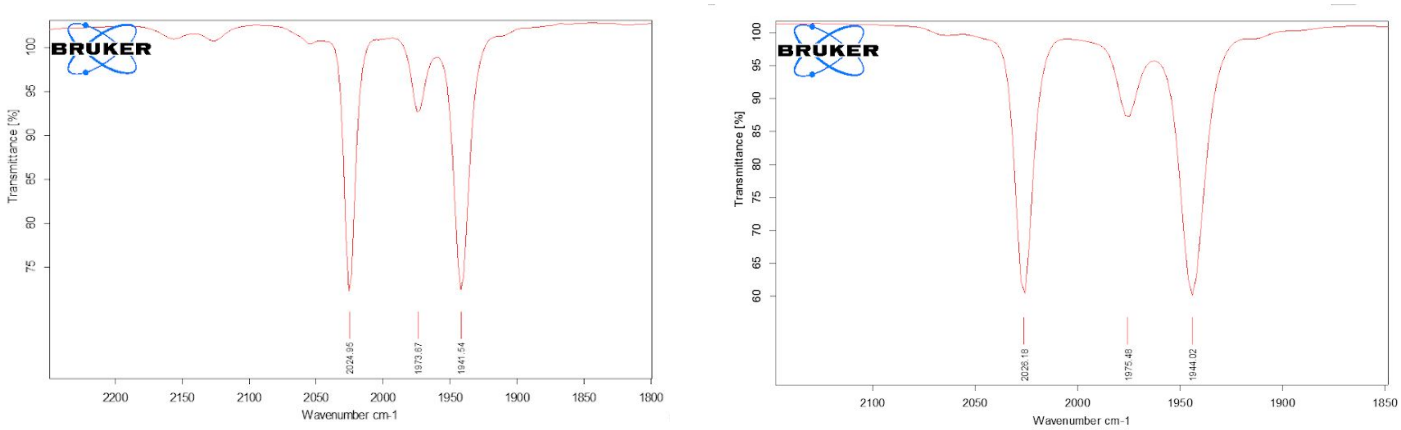
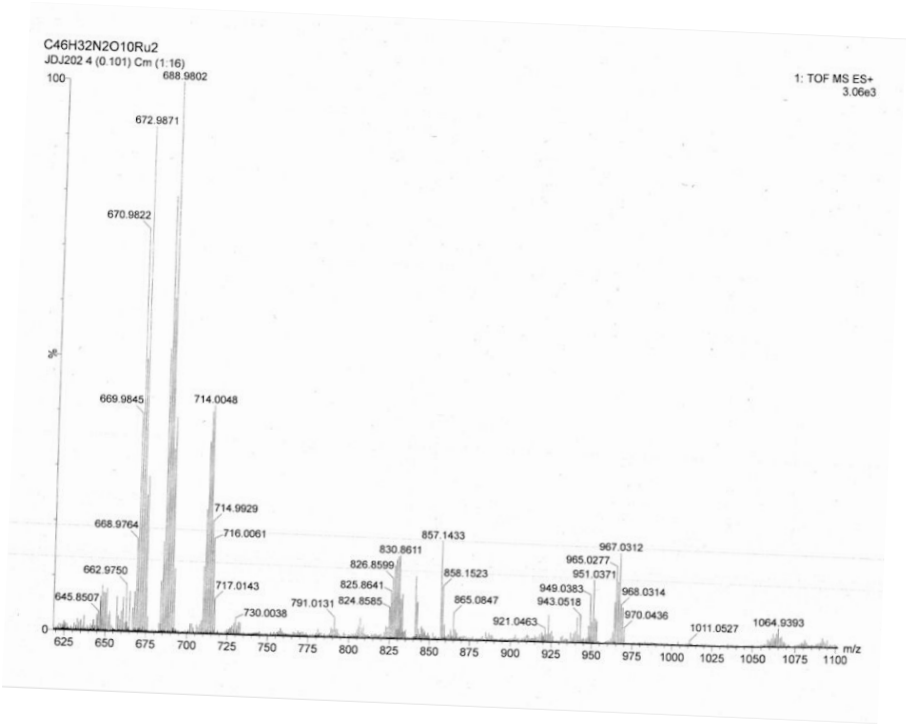


Figure 2: FTIR wave pattern of replicate sawhorse batches

We were able to synthesise 2 more batches of the sawhorse complex, both having the sawhorse structure as seen from the similar wave patterns observed from the IR graphs obtained from the two replicates (Figure 2).



We also conducted an electrospray ionization mass spectrometry (ESI-MS) to ascertain the purity of the compound synthesised. The mass spectrometry, in the case of our sawhorse complex, split a larger compound into smaller fragments. After calculation of all the peaks observed, we were able to establish the type of ligands (CO, Pyridine, Chalcone) that broke off from the larger compound during the course of electrospray mass spectrometry and therefore ascertain that the intended compound, diruthenium sawhorse complex, was successfully synthesised over the course of our experimentation.

Synthesis of Cationic complex

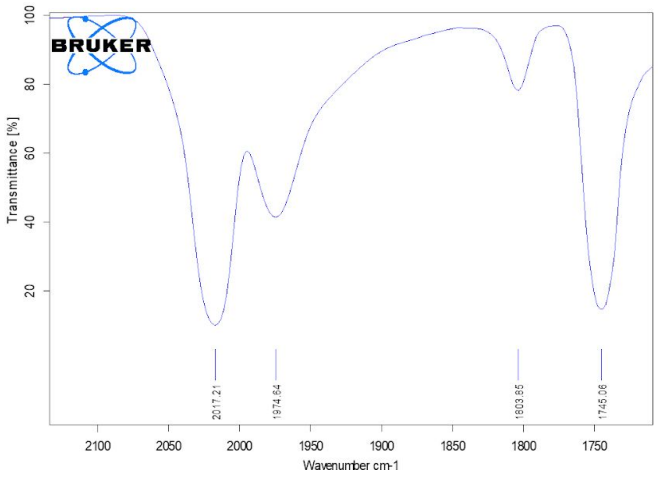
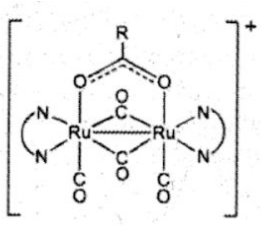


Figure 3: FTIR wave pattern of cationic complex

The cationic complex that was synthesised from the sawhorse intermediate was also analysed using both FTIR spectroscopy (Figure 3) and ESI-MS. The wave numbers emerging at 2017, 1975, 1803, 1746cm^{-1} and the wave peak pattern shown (strong-weak-weak-strong) are similar to other organometallic cationic complexes (Jiang et al., 2017), an indication that the bridging CO ligands (Figure 4, right) had been formed instead of the CO pattern found in the sawhorse structure.



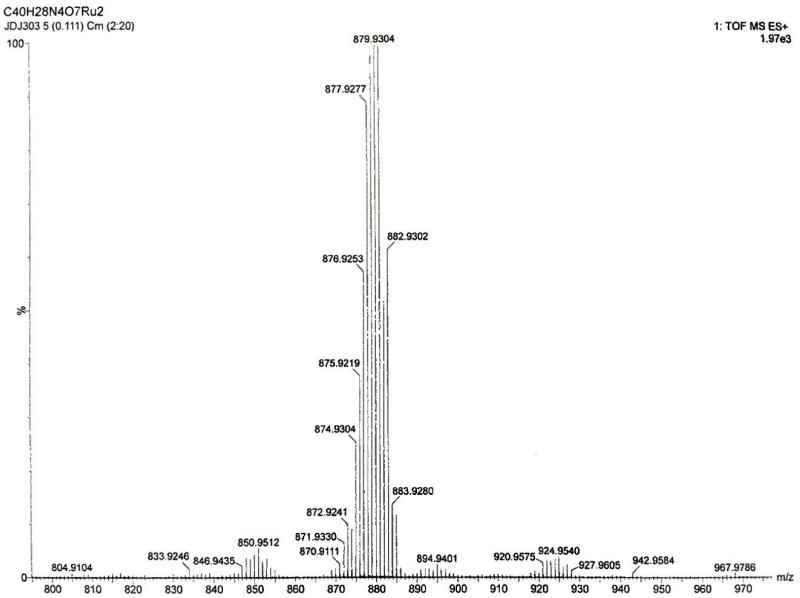


Figure 5: ESI mass spectroscopy of cationic complex

Similarly, when the compound was analysed using ESI-MS (Figure 5), the compound's purity was easier to ascertain as it was an ion. A major peak around m/z 878 was observed, which is the calculated relative molecular mass of our cationic complex. Therefore, through both FTIR as well as ESI-MS, we were able to confirm that the cationic complex had been synthesised successfully.

Conclusion

We were able to successfully synthesise our novel diruthenium sawhorse and cationic complexes incorporating carboxylated chalcone. These compounds have the potential to be used in the field of bio-imaging alongside thermography to image and identify tumours and abnormalities in the body. However, more research can be conducted on the suitability of such complexes for human usage by further examining its bioactivity in the human body, as well as expanding the scope of synthesis of novel organometallic compounds by incorporating a wider range of chalcones and its derivatives.

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