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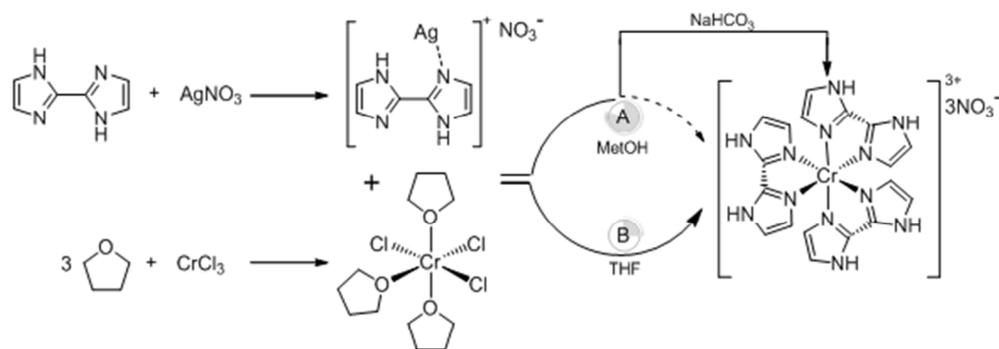
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Fast synthesis of a tris(N,N-diimine)chromium(III) complex by a microwave-assisted approach.

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Abstract

Background: In the last ten years, Photodynamic Antimicrobial Chemotherapy (PACT) has emerged as a reliable option to threat several bacterial and fungal infections. Tris(N,N-diimine)chromium(III) complexes are known for their potential as photosensitizers; however, synthesis is usually a very time consuming procedure. **Results:** With our novel methodology, $[Cr(biim)_3](NO_3)_3$ was prepared in a few seconds as a powder and the yield of the reaction was improved compared with previously described synthesis, where obtaining crystals is complicated due to the acidity of the media. The precursors for the synthesis are also synthesized through fast and simple methods. Identification of intermediates and the final product was performed by melting point, infrared, UV-vis and NMR spectroscopy. **Conclusions:** This article describe a quick, easy and eco-friendly procedure to synthesize the $[Cr(biim)_3](NO_3)_3$ employing microwave irradiation.

Graphical Abstract
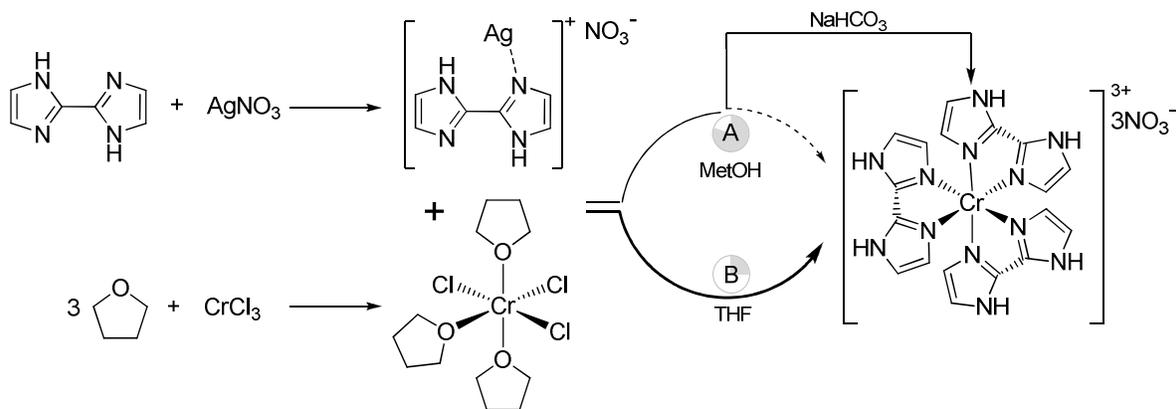
Keywords

Chromium(III) complexes, microwave, synthesis, biimidazole, Photodynamic Chemotherapy

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Background

In the last ten years, Photodynamic Antimicrobial Chemotherapy (PACT) has emerged as a reliable option to threat several bacterial and fungal infections. The search for new and better photosensitizers is an area of great interest because this therapy has proven to be a good alternative against antibiotic resistant pathogens.[1]

Different families of compounds have proved to be able to produce the death of microorganisms after irradiation at the proper wavelength, *e.g.* porphyrins, phtalocyanines, Ru(II) complexes and Cr(III) complexes.[2-5] Among these, Cr(III) complexes are the ones capable of facilitating a Type I photosensitization mechanism, producing photochemical damage without the intervention of reactive oxygen species (ROS).[6] This pathway could be an interesting alternative to some well known bacterial antioxidant mechanisms.[7] These transition metal complexes and their use in Photodynamic Therapy (PDT) and PACT has been the focus of our research for the several last years,[5,6,8,9] It have been probed that they present interesting photochemical properties to be used as antimicrobial agents; however, there are no many of them commercially available. These downfalls are, in part, due to the fact that these Cr(III) complexes are not easy to prepare and is, usually, a time consuming procedure.[10] In this work, we describe a novel and easy microwave assisted procedure to synthesize a Cr(III) complex in a few minutes. Considering that this family of complexes is known for its potential as a photosensitizer, our quick and effective synthesis would be an attractive alternative to produce useful compounds that are not currently commercially accessible.

Results and Discussion

In the synthesis of $CrCl_3 \cdot THF_3$, all the glass material and the solvent must be carefully dried. Otherwise, some hydrate will be formed and tetrahydrofuran cannot displace the water molecules once they are coordinated.

For the preparation of $[Cr(biim)_3](NO_3)_3$, a previous reported method using methanol as solvent was tried out (Scheme 1, path A).[16] Our attempts to crystallize or precipitate the complex failed. After the analysis of reaction conditions (solvent, temperature, etc.), we infer that the problem was the acidity of the media, which increases as time elapsed. HCl formed from the alcohol during reaction heating prevented the crystallization of the complex, indicating that the mechanism of coordination is not favored in the presence of H^+. Over time, the acidity of the media increased and resulted in protonation of both the little amount of complex formed and the 2,2′-biimidazole (unable to coordinate with the metal centre) (Scheme 1, paths AC). Neutralization of the crude reaction with few drops of saturated $NaHCO_3$ solution allowed for isolation of the crystalline compound (Scheme 1, paths AD).
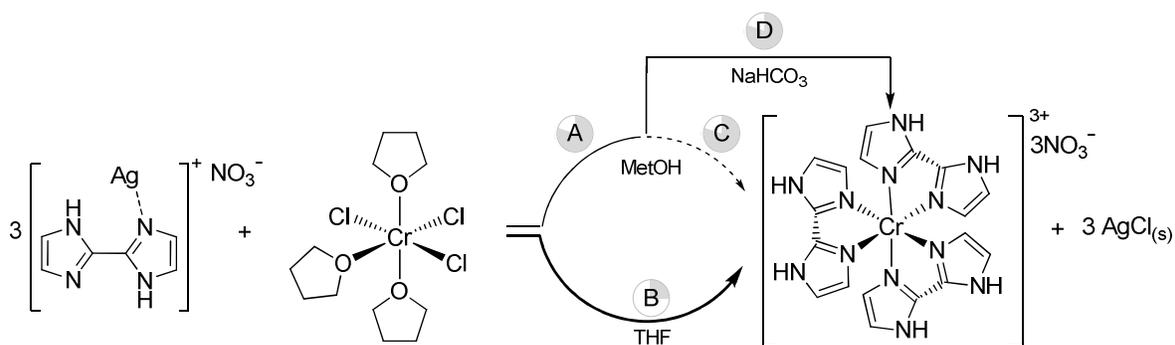
Scheme 1. Optimized synthesis of $[Cr(biim)_3](NO_3)_3$ (path AD or B) and previously reported by Gruia *et al.* (paths AC).

From these results, the synthetic procedure was modified to be carried out in dry THF and yielded off-white crystals of $[Cr(biim)_3](NO_3)_3$ after concentration (Scheme 1, path B). Optimal reaction conditions were selected after several trials at different times and

temperatures under microwave irradiation (Figure 1) and determined that the reaction reaches its maximum experimental yield (94%) after 90 seconds at 110°C. It is clear that an increase in the temperature above 110 °C is not suitable since the yield seems to decrease, possibly by thermal decomposition of the precursors or the formation of other products.
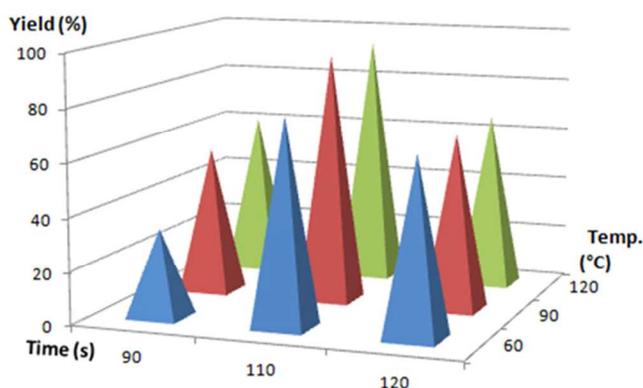
Figure 1. Time and temperature optimization for the MW induced reaction of $CrCl_3.THF_3$ and $Ag(2,2´biimidazole)NO_3$.

Experimental

Synthesis of the precursor $CrCl_3 \cdot THF_3$

Anhydrous $CrCl_3$ (1.00g, 6.31 mmol) was dissolved with the minimum amount of tetrahydrofuran (THF) in a previously dried round bottomed flask. The substitution of Cl with THF was evident from the formation of a fine-purple powder. It is important to note that the coordinated THFmolecules have ^{1}H-NMR signals that are similar to those of free THF, making differentiation between free and coordinated THF difficult. The procedure was entirely conducted inside a globe box under N_2 overpressure due to the reactivity of

the $CrCl_3$ with air humidity. The purple powder, identified as $CrCl_3 \cdot THF_3$, was obtained after drying *in vacuo* (Scheme 2).
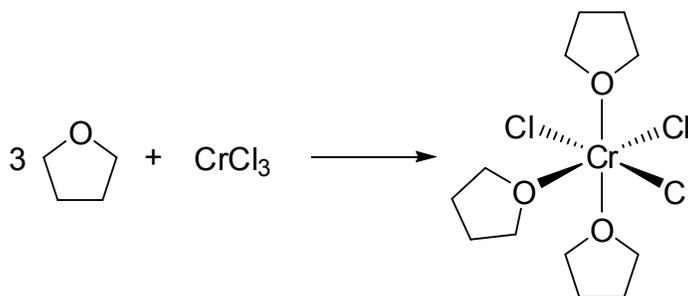
Scheme 2. Synthesis of $CrCl_3 \cdot THF_3$, building block in the synthesis of $[Cr(biim)_3](NO_3)_3$.

This novel method allows for isolation of $CrCl_3 \cdot THF_3$ in essentially quantitative yield and high purity (2.34g, 99% yield, Figure 2). This method is considerably easier and faster than the previously reported Soxhlet extraction method.[11] ^{1}H-NMR (400 MHz, acetone-d_6): 3.59 (s, broad signal, 4H); 1.80 (s, broad signal, 4H). Absorption coefficients were determined in DMSO: ε_{680}= (4.1 ± 0,1) $10^4 M^{-1}cm^{-1}$. Mp: 142.9°C. IR (KBr): 1042, 1018, 855 cm^{-1}.[12]
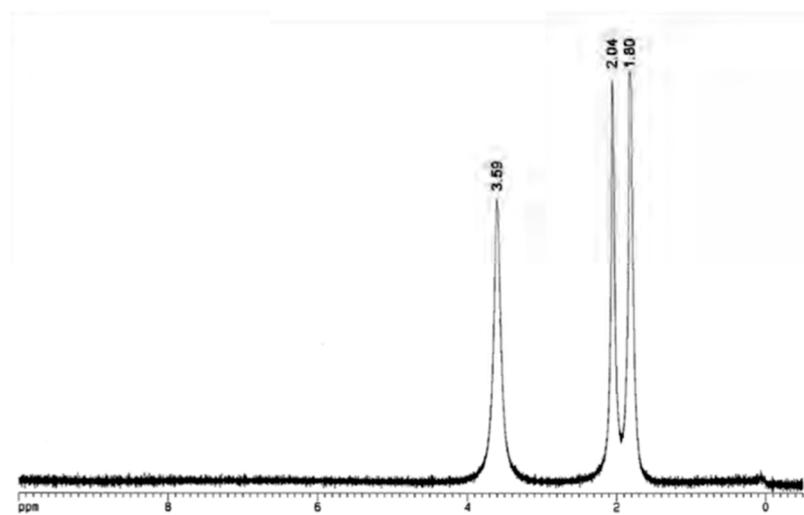
Figure 2. ^{1}H-NMR spectrum of $[CrCl_3(THF)_3]$ in acetone-d_6.

Synthesis of the precursor Ag(2,2´biimidazole)NO₃

The synthesis of Ag(2,2'biimidazole)NO$_3$ was carried out using a microwave reactor set at 300 W. Only 90 seconds at 95°C are needed to complete the reaction between the 2,2´-biimidazole (270mg, 2mmoles) and silver nitrate (340mg, 2mmoles) in nitric acid (4mL, 0.1M, Scheme 3).
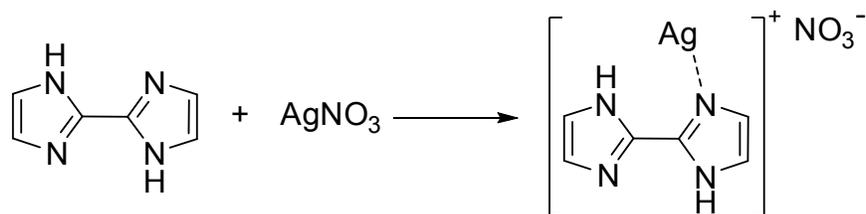
Scheme 3. Synthesis of Ag(2,2´biimidazole)NO$_3$, intermediate in the synthesis of [Cr(biim)$_3$](NO$_3$)$_3$.

Golden columnar crystals of Ag(2,2´biimidazole)NO$_3$ were formed upon evaporation of the solution with forced air flow (577mg, 95% yield, Figure 3). NMR and IR spectral analysis were in agreement with the literature.[13,14] ^{1}H-NMR (400 MHz, DMSO-d_6): 12,65 (s, broad signal, 2H); 7,06 (s, broad signal, 4H). IR (KBr): 3149, 3131, 2938, 1383 cm^{-1}.
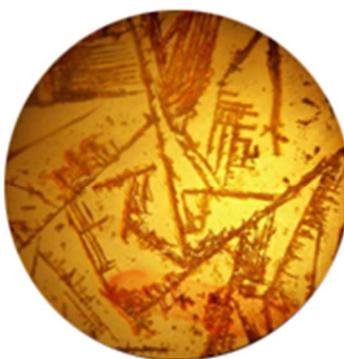
Figure 3. Ag(2,2´biimidazole)NO$_3$ crystals (2x).

Synthesis of [Cr(biim)₃](NO₃)₃

$CrCl_3.THF_3$ (32mg, 0.08mmol) and Ag(2,2′-biimidazole)NO_3 (78mg, 0.25mmol) were combined in 10mL of THF. Then the mixture was irradiated for 90 seconds after setting reaction temperature at 110°C and 300 W. After the reaction, large amounts of a white precipitate was immediately observed, due to the formation of the very insoluble AgCl. The reaction mixture was cooled and the inorganic impurity removed by filtration. Pure $[Cr(biim)_3](NO_3)_3$ was then obtained (48mg, 94% yield from THF, Scheme 1, path B) following solvent evaporation and recrystallization.

Due to the high speed and temperature required to carry out substitution reactions of the THF ligands with 3 equivalents of 2,2′-biimidazole, reaction monitoring by ^{1}H-NMR was not possible as was for Brennan *et al.* in the synthesis of analog complexes with pyridine and bipyridine. In such cases, the low temperature and high reactivity allowed the authors follow the reaction through the disappearance of the ligand signals.[15]

Initial analysis of the crude reaction by ^{1}H-NMR showed the formation of the $[CrCl_3(THF)_3]$ complex and free, uncoordinated 2,2′-biimidazole signals. After the MW reaction, disappearance of 2,2′-biimidazole signals was observed, due to its coordination with the Cr(III) centre, displacing the THF ligands. The ^{1}H-NMR (400 MHz, DMSO-d_6) spectrum of the solid clearly show traces of free 2,2′-biimidazole and broad signals corresponding to the deuterated solvent and water (Figure 4).
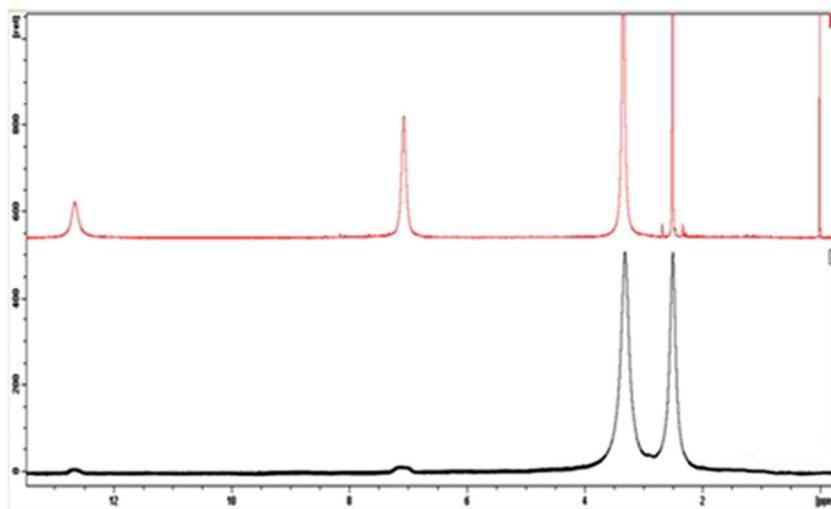
Figure 4. NMR spectra of the free ligand 2,2´-biimidazole (top) and the final complex [Cr(biim)$_3$](NO$_3$)$_3$ (bottom).

Subsequently, a new, recrystallized portion of the solid was dried and dissolved in DMSO-d_6 and a ^{13}C-NMR spectrum was recorded. The absence of any ligand resonances (2,2´-biimidazole and THF) in the resulting ^{13}C-NMR spectrum is an important result for two reasons (Figure 5). Firstly, this result is a strong indication of 2,2´-biimidazole coordination and, secondly, it suggests that the traces of 2,2´-biimidazole resonances observed in the ^{1}H-NMR (of the solid without recrystallization) are associated with free 2,2´-biimidazole. This important conclusion was also achieved by Brennan *et al.* in the synthesis of analog complexes.[15]

On the other hand, infrared analysis agrees with literature [16] and the electronic spectrum of the [Cr(biim)$_3$](NO$_3$)$_3$ shows evidence of the internal transitions of 2,2´-biimidazole: 275, 283, 288 y 296 nm, and the Cr(III)-ligand coordination: 340nm. IR (KBr): 3129, 3000 1788, 1614, 1383, 1246, 1193, 1138, 1102, 1036, 1009, 960, 943, 9224, 875, 815, 837, 763, 739, 701, 624, 539, 451 cm^{-1}. m/z: 452 [Cr(Hbiim)$_3$ + H]$^+$. The

absorption coefficients were determined in DMSO: ε_{275}= (5.23 ± 0.01) 10^4 $M^{-1}cm^{-1}$ and

ε_{340}= (2.94 ± 0.09) 10^4 $M^{-1}cm^{-1}$.
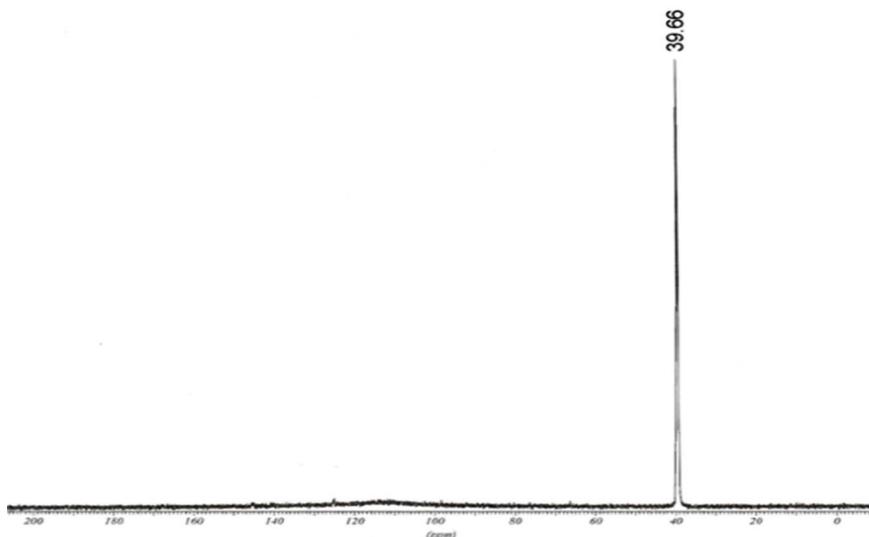
Figure 5. ^{13}C-NMR spectra of the final complex [Cr(biim)$_3$](NO$_3$)$_3$.

Conclusion

 This work has discussed a novel procedure that allows for the synthesis of [Cr(biim)$_3$](NO$_3$)$_3$ in a few seconds. This is a major advancement in the search for potentially commercial photosensitizers to be use in PDT and PACT. Currently, microbiological analyses are ongoing.

A considerably easier method to prepare CrCl$_3$•THF$_3$ was also described. This one step synthesis does not require the utilization of a time consuming Soxhlet extraction. This is important because CrCl$_3$•THF$_3$ is a commonly used precursor in several Cr(III) complex synthesis.[17] The synthesis of the Ag(2,2´biimidazole)NO$_3$ was also optimized to be carried out using a microwave reactor.

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Materials and Methods

The anhydrous $CrCl_3$ was obtained from $CrCl_3\cdot 6H_2O$ by refluxing in thionyl chloride. The 2,2´-biimidazole was purchased from MP Biomedicals and the $AgNO_3$ from Taurus, both in analytical grade. All organic solvents for the synthesis were of analytical grade and were distilled under nitrogen atmosphere prior to use.

Reactions under microwave irradiation were performed in cylindrical quartz tubes (Φ = 1.5 cm) located in a Anton Paar Monowave 300 reactor (2.455 GHz), with adjustable power within the range 0–300 W and a wave guide (monomode) fitted with a stirring device and IR and Ruby temperature detectors. Melting points were determined with an Electrothermal 9100 melting point apparatus. All compounds were characterized by standard spectroscopic techniques (NMR, UV, IR) and all data are in agreement with the proposed structures. NMR spectra were recorded in DMSO-d_6 or acetone-d_6 with a Bruker Avance II 400 MHz spectrometer (BBI probe, z gradient) (^{1}H at 400.16 MHz and ^{13}C at 100.56 MHz). Chemical shifts are reported in parts per million (ppm) downfield from TMS. The spectra were measured at 22 °C. Absorption spectra of the solutions were recorded with a UV-Vis Agilent 8243 spectrophotometer with DAD detector, using a quartz cell with an optical path length of 1 cm and acetonitrile as solvent. Infrared solid spectra were recorded with an FTIR Bruker IFS 28v spectrometer, with a resolution of 2 cm^{-1} in the range from 4000 to 400 cm^{-1} by using KBr disks.

Competing interests

The authors declare that they have no competing interests.

Authors' contribution

MJS carried out the synthesis of all compounds and its characterization. PFG helped with some of the experiments. The draft was prepared by MJS and results and discussion was guide by WJP and GAA. All authors read and approved the final manuscript.

Acknowledgements

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