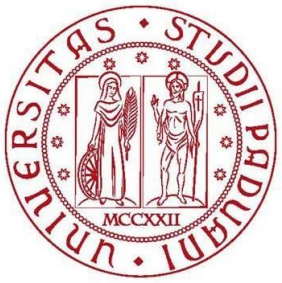

UNIVERSITA' DEGLI STUDI DI PADOVA

DIPARTIMENTO DI SCIENZE CHIMICHE

CORSO DI LAUREA TRIENNALE IN CHIMICA

APPLICATION OF CHLORO-FUNCTIONALIZED AMINOTRIPHENOLATE (TPA) COBALT COMPLEX FOR AMINES CHIROPTICAL SENSING

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SUMMARY

1 INTRODUCTION

1.1 CIRCULAR DICHROISM

Circular dichroism (CD) spectroscopy is one of the most useful techniques for the stereochemical analysis of chiral biopolymers and fine chemicals. It has become invaluable for the assignment of the absolute configuration, the study of conformational isomers, and the determination of racemization kinetics of CD active chiral compounds.[1] Molecular interactions between a nonracemic chiral substrate and a CD-silent probe (achiral compound or a racemic mixture of rapidly interconverting enantiomeric conformations) bearing a chromophoric unit can induce a strong, characteristic chiroptical readout. A covalent or noncovalent binding event that coincides with a well-defined asymmetric induction process can effectively imprint the chiral information of the substrate on the stereo-dynamic sensor and thus generate intense Cotton effects in the absorption region of the latter. The probe can thus function as a stereochemical reporter unit and analysis of the CD spectrum often provides accurate information about the absolute configuration and enantiomeric composition of the substrate used.[1] In the recent years, high-throughput screening (HTS) methods (*e.g.* parallel synthesis and combinatorial chemistry) are becoming increasingly essential in discovering chiral catalysts or auxiliaries for asymmetric transformations. These methods can lead to the exploration of a range of structural candidates and reaction conditions as a means to obtain the highest enantiomeric excess (ee) of a desired transformation. One current bottleneck in these approaches to asymmetric reactions is the determination of ee, which has led researchers to explore a wide range of HTS techniques. To be truly high-throughput, it has been proposed that a technique that can analyse a thousand or more samples per day is needed.[2] Chromatographic techniques, such as GC and HPLC, are currently the benchmark for the separation and quantification of chiral molecules. However, they still lack to be functional in the implementation for HTS of ee. This is mainly due to long elution times and, in addition, multiplex methods require specialized instrumentation. On the other hand, optical

methods, such as circular dichroism, could be a better solution because they are faster, cost-effective and they can be easily implemented into HTS.[3]

1.2 STEREO-DYNAMIC EQUILIBRIUM

In recent years the development of stereo-dynamic optical probes has been very important for their capability to act as molecular sensor for the determination of the ee of chiral compounds. In general, these molecular systems are characterized by the presence of at least one labile stereogenic element which, after the addiction of a chiral analyte, undergoes the formation of a preferential diastereoisomer (Figure 1). As a consequence, the probe is capable to recognize the chiral analyte of interest and greatly amplify its chiroptical readout. One widely used approach is based on the use of metal complexes of tetradentate ligands which assume a propeller-like rearrangement around the metal center whose P or M configuration is controlled through the incorporation of a chiral analyte. A great number of sensors have been developed, but their activity is generally restricted to one or a few classes of chemicals, and the analysis outcome relies on precise knowledge of the probe and analyte concentrations. This aspect in particular limits the potential practical applications.[4] However, the use of supramolecular sensors is increasing and is usually combined with optical signaling techniques because they are rapid and often inexpensive. For example, the tris(2-pyridylmethyl)amine (TPMA) ligand is one of the most used scaffold exploited for this purpose. Several applications of complexes based on this ligand were reported -by Canary, Anslyn, and more recently by the group where this thesis has been carried out for enantiomeric excess determination of carboxylic acids,[5] amino acids,[6,7] and alchols.[3] The chiroptical technique most employed is the Electronic Circular Dichroism (ECD) that exploits the presence of a chromophoric unit for the sensing of chiral analytes. Recently, some examples regarding the use of other chiroptical techniques were also reported due to the peculiar advantages these “alternative” techniques can offer. In 2016, an interesting application regarding the use of Vibrational Circular Dichroism (VCD) spectroscopy for the chirality recognition of several aminoacids has been reported by the group[8] followed by a study on a new fluorescent probe

based on a structural modification of the TPMA ligand. This allowed the detection of amino acids through Circular Polarized Luminescence (CPL) spectroscopy, that furnishes information on the chiroptical properties of the exited state by the inspection of the different emission of the right and left-handed circular polarized light. Similar to TPMA (Figure 1), aminotriphenolate (TPA) is characterized by a propeller-like arrangement of the ligand around the metal center when viewed along the metal−nitrogen axis. As a consequence, C_3-symmetric trigonal bipyramidal (TBP) complexes are obtained in the two helical arrangements, which interconvert at room temperature, yielding a racemic mixture of enantiomeric complexes.[4]

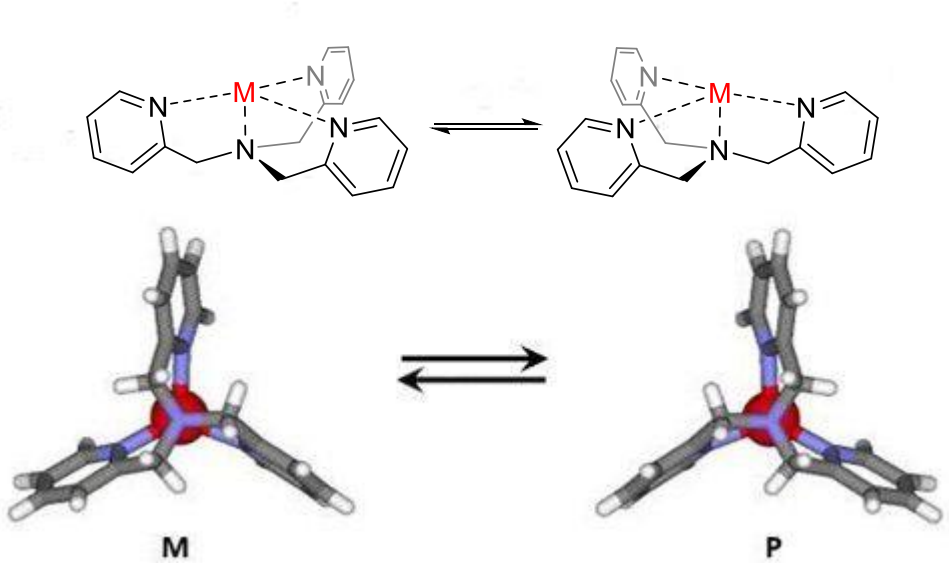


Figure 1. Clockwise (P) and counter-clockwise (M) arrangement of **TPMA** complexes.

Also with these systems, a relevant example where an oxo-vanadium TPA complex **1**. TPA complexes were developed as a probe able to gather ee without knowing the concentrations of the different species in solution has been reported by the group where this thesis has been carried out. This complex revealed the capability to coordinate a variety of substrates giving excellent CD response. Additionally, the complex is easily prepared by mixing the opportune triphenolamine ligand and an equimolar amount of vanadium (V) oxytriisopropoxide.[4]

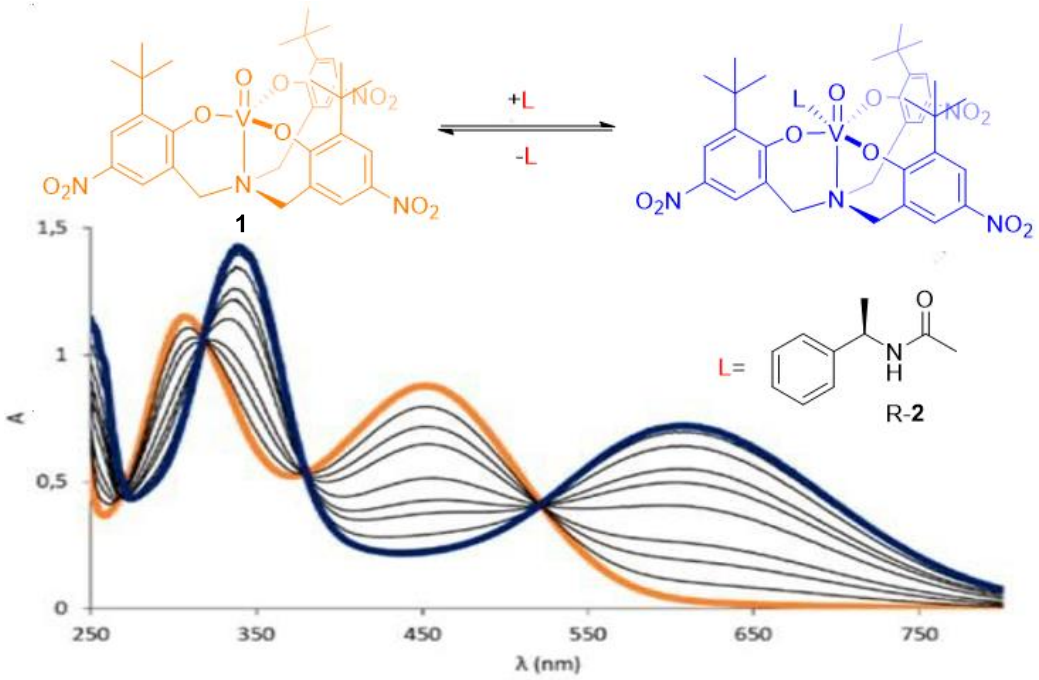


Figure 2. Oxo-vanadium TPA complex (**1**) and an enantiopure amide R-**2**; UV titration of a 5.0 x 10^{-5} M $CHCl_3$ solution of **1** with compound R-**2**. The spectrum of pure **1** (bold orange line) and that in the presence of 20 equiv of R-**2** (bold blu line) are highlighted.

Complex **1** shows strong absorptions in the UV/vis spectrum, and two bands are observed at 308 and 450 nm (Figure 2). This is due to ligand-to-metal charge transfer (LMCT) from the phenolate oxygen to the vanadium(V) empty d orbitals. Afterwards, when the complex **1** is dissolved in common coordinating solvents or in the presence of a donor species its UV/vis spectrum exhibits red-shifts of both bands. Specifically, the second band moves significantly up from 450 to 600 nm, reflected in a marked color change of the solution from orange to deep blue. The confirmation of the binding was also achieved in the solid state through X-ray analysis of suitable crystals obtained from a toluene solution of **1** in the presence of the enantiopure amide S-**2** (Figure 3).

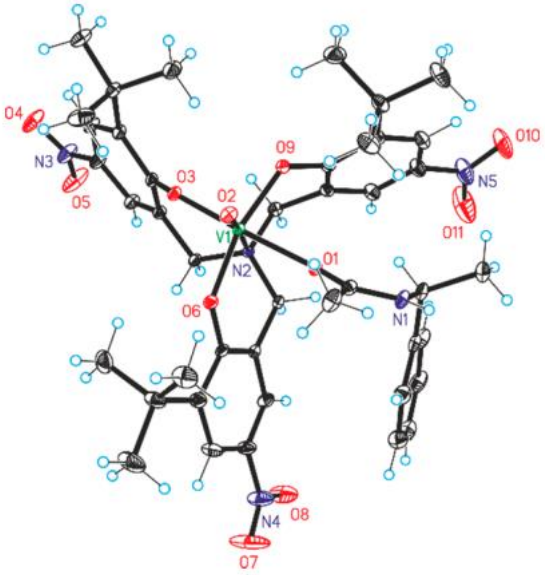


Figure 3. ORTEP representation of the adduct between complex **1** and amide S-**2**.

The CD spectrum of a chloroform solution of **1** in the presence of amide R-**2** showed strong Cotton effects in the visible region between 600 and 350 nm (Figure 4a). No dichroic signal was observed in the same region when CD analysis was performed on either pure complex **1** or amide guest R-**2**. A mirror image spectrum was obtained when S-**2** was used instead of R-**2** in the presence of **1**. The same CD pattern was also observed with chiral primary amines **3**−**4** (Figure 4b-c). The sign of CD bands for primary amines revealed a common trend since in all case negative signals were observed with R compounds and vice versa.

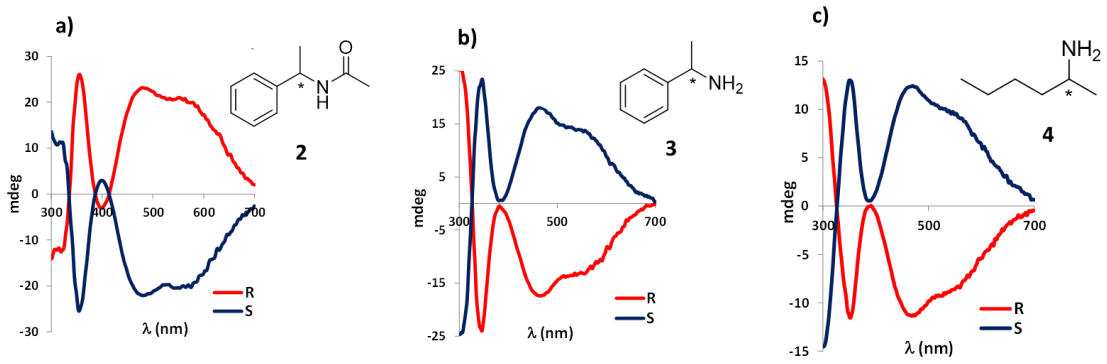


Figure 4. CD spectra recorded between 700 and 300 nm for chloroform solutions of complex **1** (10^{-4} M) in the presence of different chiral compounds: (a) [**2**] = 2 x 10^{-4} M, (b) [**3**] = 2 x 10^{-3} M, (c) [**4**] = 5 x 10^{-4} M.

1.3 ANISOTROPIC g-FACTOR

In order to test whether **1** could be exploited for ee determination, a number of measures were conducted on the CD ellipticity values (θ) for a series of samples containing **1** and R- or S-**2** at known enantiopurity (Figure 5a). A straight linear relationship was observed between the CD response and the ee of **2** (Figure 5b).

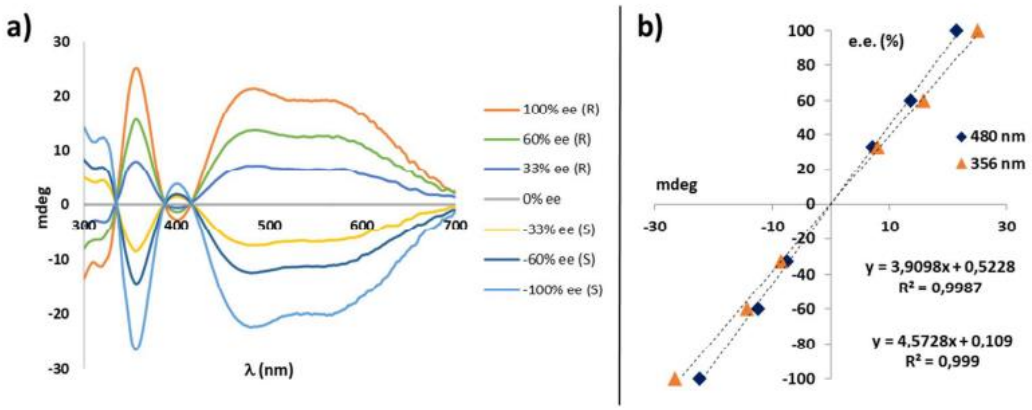


Figure 5. a) CD spectra of complex **1** in the presence of amide **2** at different enantiopurities; b) Calibration curves of amide **2** ee with respect to the ellipticity values at 480 nm and 356 nm.

However, while by knowing analyte and probe concentrations it is possible to know with a good accuracy the ee, it has been reckoned that the probe theoretically has the possibility to identify the ee independently of the concentrations of the two partners, due to the unique properties of the vanadium TPA complex to change absorbance upon coordination of the analyte. In particular, CD detector has the ability to record simultaneously both dichroic signal (Δε) and absorbance (ε), it is possible to directly find the anisotropic g-factor (g=Δε/ε). This parameter is independent from the concentration of the octahedral species but proportional to the ee. Although the g-factor has been successfully employed for stereochemical analyses with chromatographic methods, this is the first example of application for ee determination with stereo-dynamic probes. This is due to the difficulty in locating a spectral region where only the CD-active diastereomeric adduct absorbs.[4] In the case of the TPA vanadium system **1**, only the octahedral

species is responsible for the light absorption at wavelengths >600 nm (Figure 2), thus permitting the use of the anisotropic g-factor.[4]

1.4 TOWARD A NOVEL METAL

In the quest to extend the versatility of TPA systems, we performed a bibliographic search about the possibility to use different metals capable to bind novel analytes. With this perspective, we reckon that cobalt (II) was forming anionic complexes in the presence of a base. In particular, in a work by Kleij[9] an amine was acting as counter anion for a TPA cobalt (II) complexe. It should be noted that in the reported case the amine was achiral but interacting with the metal center.

2 AIM OF THE THESIS

The aim of the thesis is the development of a new stereo-dynamic probe for amines based on a chloro-functionalized aminotriphenolate (TPA) cobalt (II) anionic complex.

In particular, the experiments will be directed toward to: i) the preparation of anionic TPA complexes with (a)chiral amines, ii) the characterization of the complexes, iii) the investigation of its chiroptical properties.

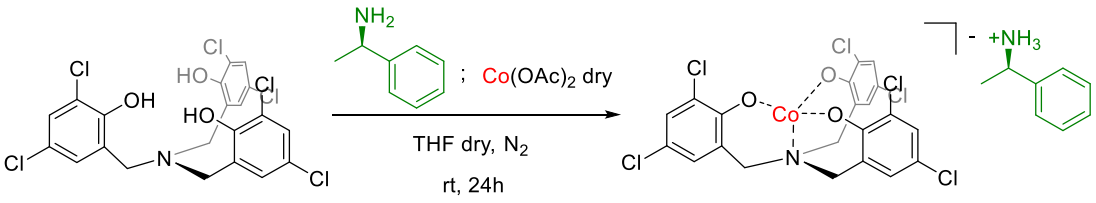
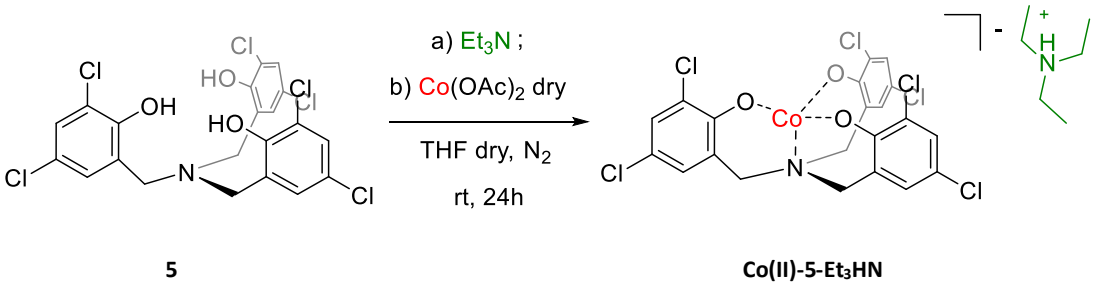


Figure 6. Synthesis of chloro-functionalized aminotriphenolate cobalt (II) with (R)-methylbenzylamine.

3 RESULTS AND DISCUSSION

3.1 SYNTHESIS OF TPA Co (II) TPA COMPLEX WITH TRIETHYLAMINE

Our initial interest was directed toward the test of the capability to form anionic complexes in presence of an organic amine. Two steps were performed trying to obtain the desired complex. Firstly, one equivalent of TPA ligand **5** was deprotonated by three equivalents of triethylamine. After 5 min, dry cobalt (II) acetate was added as the metal source for the formation of the complex (Scheme 1).



Scheme 1. Synthesis of the cobalt-TPA complex **Co(II)-5-Et$_3$HN** using dry cobalt (II) acetate in THF under inert atmosphere.

Regarding the ligand **5**, it was previously synthetized by the group where this thesis was carried out and it was already available in the laboratory. The reaction was performed in THF under N_2 and anhydrous condition. After 24 hours, the solution turned deep red and the formation of the complex was then confirmed by ESI-MS spectroscopy of the solution that shows the formation of the anionic metal species (Figure 7).

Crystallization attempts were performed once the reaction was completed using different co-solvents (hexanes, diethylether, …). Unfortunately, even though many attempts varying several parameters were carried out, it was not possible to obtain a solid precipitate.

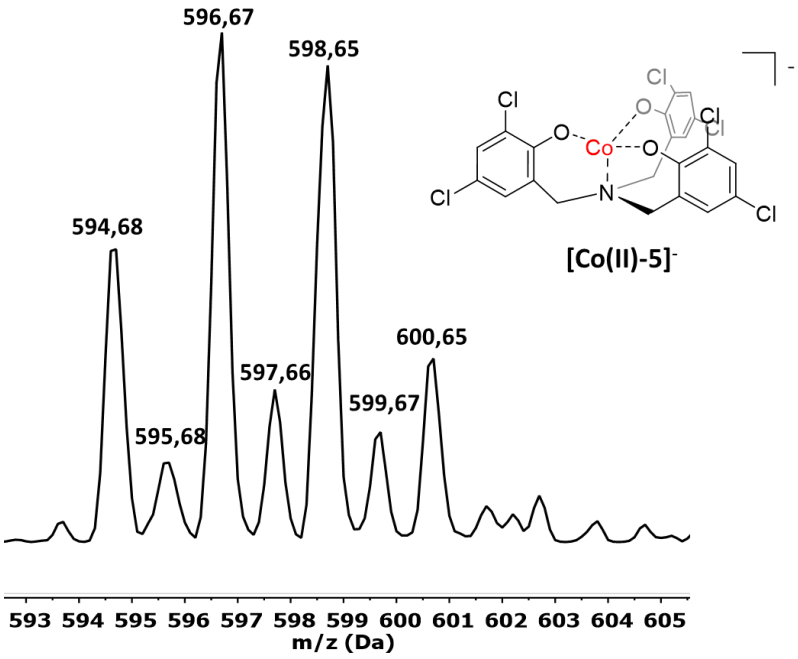
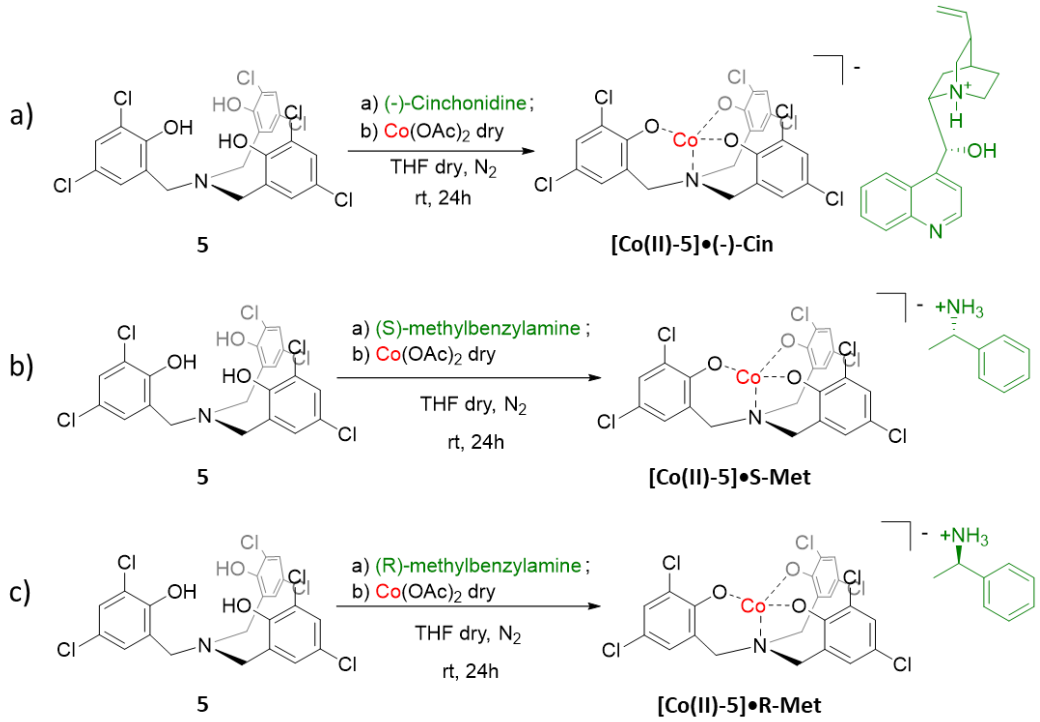


Figure 7. Experimental ESI-MS of protonated anionic complex [**Co(II)-5**]$^-$ at 596,7 m/z in ACN/H$^+$.

3.2 SYNTHESIS OF TPA Co (II) TPA COMPLEX WITH CHIRAL AMINES

The formation of the anionic cobalt complex with the triethylamine prompted us to test different amines for this synthesis. In particular, two chiral amines were tested to analyze the capability of the complex to recognize them through chiroptical investigation. Cinchonidine and methylbenzylamine were chosen as commercially available, for the latter both the enantiomers were available and thus tested for the complex formation (Figure 7). Using the same conditions as for the achiral amine, the complex formation was carried out for the three amines and the solutions analysed using ESI-MS analysis. In all the cases, formation of the anionic complex was revealed.



Scheme 2. Synthesis of anionic cobalt-TPA complex using different chiral amines, respectively *a)* (-)-Cinchonidine; *b)* (S)-methylbenzylamine; and *c)* (R)-methylbenzylamine.

3.3 ANALYSIS OF THE DICHROIC SIGNAL

As mentioned before, main purpose of this work is to verify the capability of the anionic complex **[Co(II)-5]**$^-$ to act as a stereodynamic probe for chiral amines. More in detail, the work will investigate if the interaction between the anionic complex and the protonated amine is enough to induce a preferential helical conformation of the complex to give a chiroptical readout. To verify this, the dark red solution obtained after the synthesis was analyzed with CD spectroscopy, using a 1 mm cuvette path length. The crude solution was analysed without dilutions and Figure 8 shows the CD spectra collected within the range 350 – 650 nm. For all the three amines tested it was possible to observe a good dichroic response, with a signal around 10 mdeg for all the substrates. Moreover, the dichroic signal recorded for the two enantiomers (R) and (S)-methylbenzylamine showed a good mirror image profile, indicating the opposite helicity induction of the anionic cobalt complexes. These results strongly suggest the interaction of the ammonium ion with the anionic complex.

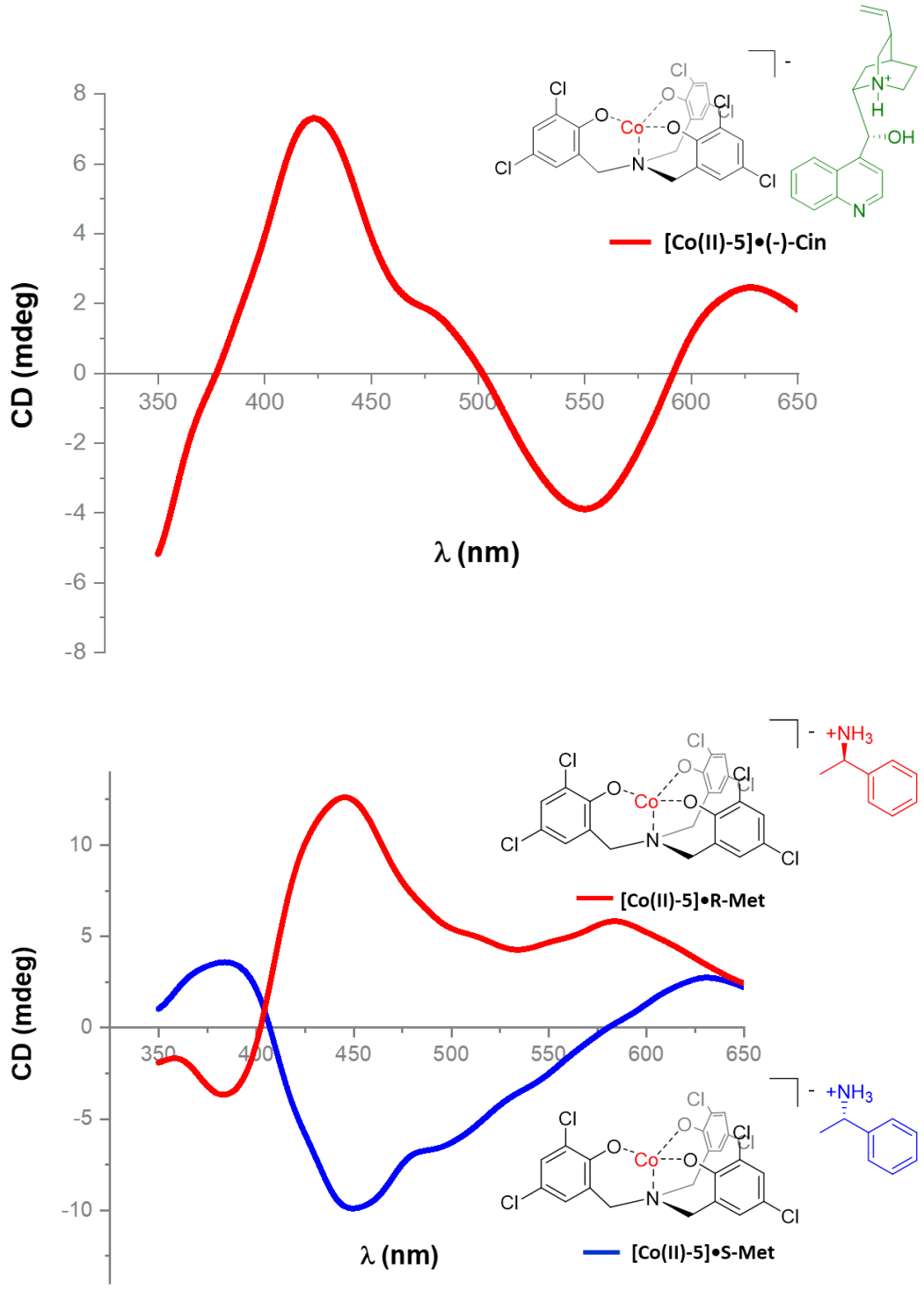


Figure 8. CD spectra within the range 350 – 650 nm of the anionic cobalt complex **[Co(II)-5]**$^-$ coordinating different chiral amines, the (-)-Cinchonidine (top) and the (R)/(S)-methylbenzylamine (bottom). The experimental parameters adopted for the CD spectra acquisition were: range 350 - 650 nm, CD scale 200 mdeg/1.0 dDD, D.I.T. 1 s, data pitch 0.2 nm, scanning speed 100 nm/min.

4 CONCLUSIONS

The studies carried out during this thesis have shown the possibility to form an anionic cobalt (II) TPA complex using an amine. Moreover, the research has also shown the capability of the anionic cobalt complex to interact with the chiral ammonium salt formed acting as a stereodynamic probe.

Future work will be dedicated to: i) the ampliation of the number of substrates, ii) the exploration of the structural influence of the substate on the chiroptical signal and iii) to a further characterisation on the type of interaction responsible for the observed signals. The recognition mechanism will be also investigated through computational and crystallographic methods.

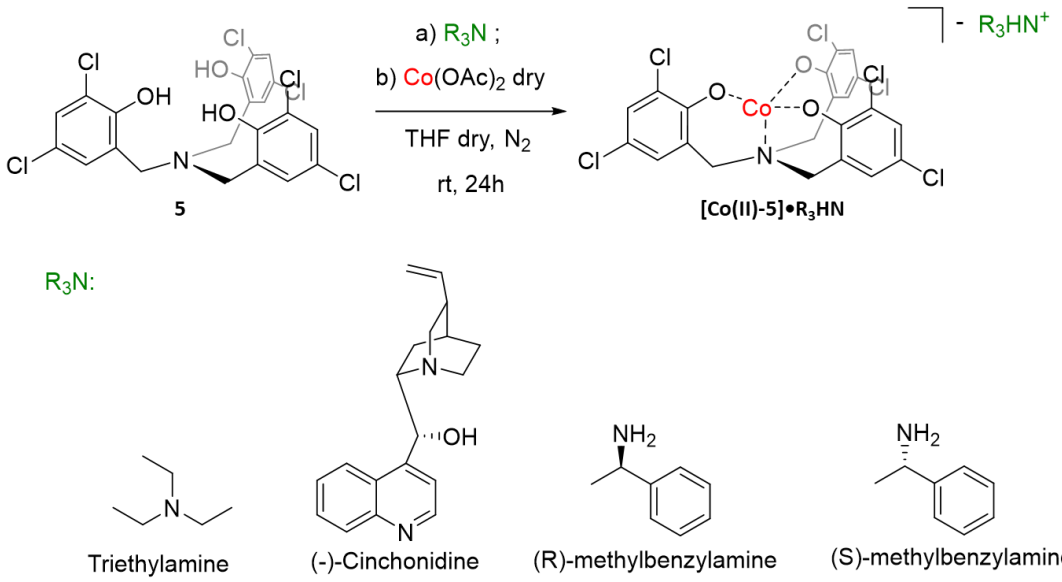
5 EXPERIMENTAL SECTION

ESI-MS spectra have been acquired with an Agilent Technology LC/MSD Trap SL, interfaced to an Agilent 1100 binary pump. The samples were preventively diluted in acetonitrile and then injected via direct infusion with a syringe pump at a rate of 0.3 ml/min. MS peak intensity for each analysis is reported as monoisotopic mass and the data were processed with Data Explorer 4.2 or with MestReNova 12.0.2. The isotopic distribution simulations have been performed through www.envipat.eawag.ch.

ECD spectra were recorded with a Jasco J-1500 spectrometer and processed with Spectra Manager Version 2.15.3.1 or OriginPro 2018 (64-bit) SR1 b9.5.1.195.

Chemicals were purchased from Aldrich, TCI, or Apollo Scientific and used without further purification.

5.1 GENERAL METHOD OF SYNTHESIS OF COBALT (II) AMINOTRIPHENOLATE COMPLEXES



Scheme 3. General synthetic procedure for the synthesis of the anionic cobalt-TPA complex using different amines.

In a balloon flask 7,4 mg of ligand **5** was introduced and dissolved in 2,5 mL of THF, obtaining a red-brown solution. Then, three equivalents of the amine (R_3N) were added in order to deprotonate the ligand. After 5 min, 2,3 mg of dry

$Co(OAc)_2$ were added using a small amount of THF. The reaction was left under stirring for 24 h, obtaining the complex **Co(II)-5-R_3HN.**

Amine	Ligand mg (mmol)	RNH mg (mmol)	$Co(OAc)_2$ mg (mmol)
Triethylamine	7.3 (0.0134)	5.1 (0.0504)	2.4 (0.0134)
(-)-Cinchonidine	7.5 (0.0138)	12.1 (0.041)	2.7 (0.015)
(S)-(-)-α-methylbenzylamine	7.4 (0.0136)	11.5 (0.0945)	2.3 (0.0132)
(R)-(+)-α-methylbenzylamine	7.1 (0.0131)	9.8 (0.0809)	2.3 (0.0131)

Table 1. Amount of the reagents used for the synthesis of the Co(II)TPA complexes

ESI-MS spectra were registered as to verify the formation of the complex. A small quantity of the mixture once the reaction was finished was dissolved in acetonitrile. ESI-MS spectra were registered in negative mode in order to detect the complex and in positive as to verify the competition of the reaction.

ESI-MS (m/z): calculated for [**Co(II)-5**]$^-$ 596.8; experimental 596,7

6 REFERENCES

1. C. Wolf, and K. W. Bentley, *Chem. Soc. Rev.*, **2013**, 42, 5408-5424.
2. D. Leung; S. O. Kang, and E. V. Anslyn, Chem. Soc. Rev., **2012**, 41, 448-479.
3. L. You; G. Pescitelli; E. V. Anslyn; L. Di Bari, *J. Am. Chem. Soc*. **2012**, 134, 7117–7125.
4. P. Zardi; K. Wurst; G. Licini, and C. Zonta, *J. Am. Chem. Soc*. **2017,** 139, 15616-15619.
5. L. A. Joyce; M. S. Maynor; J. M. Dragna; G. M. Da Cruz; V. M. Lynch; J. W. Canary; E. V. Anslyn; *J. Am. Chem. Soc*. **2011**, 133, 13746–13752
6. F. A. Scaramuzzo; G. Licini, and C. Zonta, *Chem. Eur*. J., **2013**, 19, 16809- 16813.
7. L. A. Joyce; J. W. Canary, and E. V. Anslyn, *Chem. - A Eur. J*. **2012**, 18, 8064–8069.
8. R. Berardozzi; E. Badetti; N. A. Carmo dos Santos; K. Wurst; G. Licini; G. Pescitelli; C. Zonta, and L. Di Bari, *Chem. Commun*., **2016**, 52, 8428
9. C. Martín, C. J. Whiteoak, E. Martin, E. C. Escudero-Adan, J. R. Galan-Mascaros, A. W. Kleij, *Inorg. Chem.* **2014**, 53, 11675−11681