Cobalt picolinamide complexes as potential anti-cancer agents

Laura Hester Dena Ghandhi

Submitted in accordance with the requirements for the degree of
Doctor of Philosophy

The University of Leeds
School of Chemistry

April, 2017

The candidate confirms that the work submitted is his/her own and that appropriate credit has been given where reference has been made to the work of others.

This copy has been supplied on the understanding that it is copyright material and that no quotation from the thesis may be published without proper acknowledgement.

The right of Laura Hester Dena Ghandhi to be identified as Author of this work has been asserted by her in accordance with the Copyright, Designs and Patents Act 1988.

Acknowledgements

Abstract

This thesis details the synthesis of cobalt picolinamide complexes with potential chemotherapeutic applications. The anti-cancer, anti-bacterial and anti-fungal activity of these complexes was probed, with lead complexes undergoing further mechanistic investigations.

Three series of cobalt picolinamide complex were investigated: cobalt(III) *tris*-picolinamide, cobalt(II) *bis*-picolinamide and cobalt(III) mixed ligand complexes. Cobalt *tris*-picolinamide complexes consist of a cobalt(III) ion surrounded by three picolinamide ligands, bound through the pyridyl and amide nitrogen atoms. A minor isomer with different ligand coordination was formed under certain reaction conditions, provided that an electron donating group is present as a substituent on the picolinamide ligand. The formation of cobalt *bis*-picolinamide complexes was also only successful when electron donating groups were present on the picolinamide ligand. These complexes contain a cobalt(II) ion with two picolinamide ligands and two axial thiocyanate ligands. The *cis/trans* orientation of the thiocyanate ligands varies dependent upon the position of the picolinamide ligand substituent. Mixed ligand complexes consist of a cobalt(III) ion with two picolinamide ligands and one β-diketonate or ferrocenyl β-diketonate ligand.

Complexes were screened for their anti-cancer potential against a number of cell lines. Cobalt *bis*-picolinamide and mixed ligand complexes were non-toxic. Some cobalt *tris*-picolinamide complexes displayed cytotoxicity, with the minor isomer displaying greater activity than the analogous major isomer. The two lead complexes were active against cancer cells and cancer stem cells. The mechanism of action is proposed to be inhibition of cell proliferation through interruption of the cell cycle at M phase. The lead complexes did not undergo hydrolysis, in contrast to the mixed ligand complexes containing the ferrocenyl β-diketonate ligand. The lead complexes could also adsorb onto an artificial biomembrane surface, unlike the inactive complexes, implying a correlation between cytotoxicity and cellular uptake. Additionally, cobalt *bis*-picolinamide complexes displayed anti-fungal activity against *C. albicans*, with the thiocyanate ligands essential for activity.

Table of Contents

List of Abbreviations

°	degrees
δ	chemical shift
η	hapticity
λ	wavelength
μ_B	Bohr magnetons
μg	microgram
μL	microlitre
μM	micromolar
μm	micrometre
Å	Angstrom
A	adenine
acac	acetylacetonate
ADR	adverse drug reaction
ARPE-19	human retinal pigment epithelial cell line
azaCBI	azachloromethylbenzindoline
BC	Before Christ
bca	*bis*(2-chloroethyl)amine
bce	N,N'-*bis*(2-chloroethyl)ethylenediamine
BE	human colon carcinoma cell line
bpy	2,2'-bipyridine
br. s	broad singlet
°C	degrees Celsius
CAMHB	cation adjusted Mueller Hinton broth
CC_{50}	concentration for 50 % cell death
CFU	colony-forming units
cm	centimetre
CO-ADD	Community for Open Antimicrobial Drug Discovery

COD	*cis*-1,5-cyclooctadiene
CORM	carbon monoxide releasing molecule
COX	cyclooxygenase
Cp	cyclopentadiene
Cp*	pentamethylcyclopentadiene
CSC	cancer stem cell
CT	computed tomography
cyclam	1, 4, 8, 11-tetraazacyclotetradecane
cyclen	1, 4, 7, 10-tetraazacyclododecane
d	deuterated
d	doublet
ddd	doublet of doublet of doublets
Da	Dalton
DAPI	4',6-diamidino-2-phenylindole
dce	N,N-*bis*(2-chloroethyl)ethylenediamine
DFT	density functional theory
DMEM	Dulbecco's modified eagle medium
DMSO	dimethyl sulphoxide
DNA	deoxyridonucleic acid
DOPC	dioleoyl phosphatidylcholine
EDTA	ethylenediaminetetraacetic acid
e.g.	*exempli gratia*, for example
en	ethylenediamine
ESI	electrospray
et al.	and others
FBS	foetal bovine serum
FDA	Food and Drug Administration
g	gram
G	Gauss

G	guanine
h	hours
HBSS	Hank's balanced salt solution
HEK	human embryonic kidney cell line
HIF-1	hypoxia inducible factor-1
HMLER	human mammary epithelial cell line
HMLER-shEcad	human mammary epithelial cell line (CSC-enriched)
HPV	Human papilloma virus
HRE	hypoxia responsive element
IC_{50}	concentration for 50 % growth inhibition
ICP-MS	inductively coupled plasma mass spectrometry
i.e.	*id est*, that is
IGF	insulin-like growth factor
IR	infrared
in vacuo	under vacuum
in vitro	in glass
in vivo	in a living organism
J	coupling constant
K	degrees Kelvin
keto	β-diketonate
kG	kilo Gauss
log *P*	partition coefficient
m	multiplet
MHz	mega Hertz
MIA PaCAa-2	human pancreatic carcinoma cell line
MIC	minimum inhibitory concentration
min	minute
mL	millilitre
mM	millimolar

mm	millimetre
mmol	millimole
MMP	matrix metalloproteinase
MS	mass spectrometry
MTT	3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide
mV	millivolt
NAD(H)	nicotinamide adenine dinucleotide
NADP(H)	nicotinamide adenine dinucleotide phosphate
NBS	non-binding surface
NHS	National Health Service
NIR	near infrared
nm	nanometre
NMR	nuclear magnetic resonance
NSAID	nonsteroidal anti-inflammatory drug
NSCLC	non-small cell lung cancer
PARP	poly ADP ribose polymerase
PBS	phosphate buffered saline
PET	positron emission tomography
phen	phenanthroline
pK_a	acid dissociation constant
PKC	protein kinase C
pica	picolinamide
ppm	parts per million
pta	1,3,5-triaza-7-phosphaadamantane
RCV	rapid cyclic voltagram
RNA	ribonucleic acid
ROS	reactive oxygen species
s	second
s	singlet

SAR	structure activity relationship
SCO	spin crossover
sh	shoulder
SPECT	single-photon emission computed tomography
SQUID	superconducting quantum interference device
t	triplet
td	triplet of doublets
TLC	thin layer chromatography
TNFα	tumour necrosis factor-α
tpa	*tris*(2-methylpyridyl)amine
U87	human glioblastoma cell line
UK	United Kingdom
UV	ultraviolet
UV/vis	ultraviolet-visible
V	volt
VEGF	vascular endothelial growth factor
via	by way of
YPD	yeast extract-peptone dextrose

Glossary

Anaemia: decrease in the total amount of red blood cells or haemoglobin in the blood

Angiogenesis: the formation of new blood vessels

Apoptosis: the process of programmed cell death

Autophagy: destructive mechanism of the cell that disassembles unnecessary or dysfunctional components

Biofilm: any group of microorganisms in which cells adhere to each other and to a solid surface

Carcinogenesis: the initiation of cancer formation

Carcinoma: a cancer arising in the epithelial tissue of the skin or of the lining of the internal organs

Chromosome: a thread-like structure of nucleic acids and protein found in the nucleus, carrying genetic information in the form of genes

Cytosol: the aqueous component of the cytoplasm of a cell

Emesis: the action or process of vomiting

Eukaryote: an organism with a complex cell or cells, in which the genetic material is organised into a clearly defined membrane-bound nucleus

Genome: the genetic material of an organism

Homeostasis: the tendency towards a relatively stable equilibrium between interdependent elements, especially as maintained by physiological processes

Hypoxia: deficiency in the amount of oxygen reaching the tissues

Iatrogenic: relating to illness caused by medical examination or treatment

Infarction: obstruction of the blood supply to an organ or region of tissue, typically by a thrombus or embolus, causing local death of the tissue

Intercalation: insertion of a molecule into DNA between the bases

Kinase: an enzyme that catalyses the transfer of phosphate groups from high-energy, phosphate-donating molecules to specific substrates

Leukopenia: a decrease in the number of white blood cells (leukocytes) in the blood

Lymphoma: a cancer of the lymphatic system

Metastasis: the spread of a cancer or other disease from one organ or part of the body to another

Mitochondria: an organelle in which the biochemical processes of respiration and energy production occur

Mitosis: a part of the cell cycle when replicated chromosomes are separated into two new nuclei

Morbidity: the condition of being diseased

Mutation: a change that occurs in the DNA sequence

Nephrotoxicity: toxicity to the kidneys

Neurotoxicity: toxicity to the nervous system

Normoxia: normal levels of oxygen reaching the tissues

Oncogene: a gene that has the potential to cause cancer

Ototoxicity: toxicity to the ear leading to hearing loss

Paresthesia: an abnormal sensation such as tingling, tickling, pricking, numbness or burning of the skin with no apparent physical cause

Polyp: a small growth, usually benign and with a stalk, protruding from a mucous membrane

Protease: any enzyme that performs proteolysis by hydrolysis of peptide bonds

Proteasome: protein complexes which degrade unneeded or damaged proteins by proteolysis

Replication: the biological process of producing two identical replicas of DNA from one original DNA molecule

Telomerase: a protein which adds a repeat sequence to the end of telomeres

Telomere: a region of repetitive nucleotide sequences at each end of a chromosome

Transcription: the first step of gene expression, in which a particular segment of DNA is copied into RNA

Ubiquitination: the addition of ubiquitin to a substrate protein to signal degradation by the proteasome

Chapter 1
Introduction

1. Introduction

1.1 Cancer

A frequently quoted statistic is that one in two people in the UK will develop cancer in their lifetime.[1] Cancer is the leading cause of death in the UK, with 163,444 deaths in 2014 (greater than 25 % of the total number of deaths)[2] and an annual NHS spend of over £ 5 billion (6 % of total NHS budget).[3] There were 356,860 new cases in 2014 and despite there being over 100 different types of cancer, over half of new cases are accounted for by lung, breast, prostate and colorectal cancers.[4] Worldwide, cancer is estimated to be the cause of a total of 13 % of all deaths with the majority of these occurring in low- or middle-income countries.[5-7] As the global population grows and ages, the burden of cancer is likely to become ever more significant.

Cancer is an umbrella term for a heterogeneous group of disorders which are characterised by uncontrollable cell division and tissue invasion. The "multiple-hit model" states that the transformation of a normal cell to a cancerous cell (carcinogenesis) results from the accumulation of harmful genetic mutations.[8] Mutations can be inherited or, in the vast majority of cases, acquired. Acquired mutations may result from physical factors such as UV and ionising radiation, chemical factors such as tobacco and asbestos or biological factors such as infection with HPV.[9] A number of DNA repair processes exist to remove mutations. However, these are not 100 % efficient and indeed efficiency of DNA repair decreases with age and consequently there is greater potential for the accumulation of mutations.

In carcinogenesis, there are six essential cellular processes which are subverted through mutation; collectively these acquired capabilities dictate malignant growth (**Table 1.1**). The mechanism by which these capabilities are acquired, and their chronology, will differ between malignancies. The subversion of these physiological processes is possible due to two enabling characteristics: genomic instability and tumour-promoting inflammation. Emergent processes, which may be recognised as essential in the future, are deregulation of cellular energetics and evasion of immune destruction.[10, 11]

Table 1.1: Six cellular processes of carcinogenesis[10, 11]

Acquired capability	Example of mechanism
Limitless replicative potential	Activation of telomerase
Self-sufficiency in growth signals	Activation of *ras* oncogene
Insensitivity to growth inhibition signals	Deactivation of pRb tumour suppressor
Evasion of apoptosis	Production of IGF survival factors
Sustained angiogenesis	Production of VEGF inducer
Tissue invasion and metastasis	Inactivation of E-cadherin

Cancer produces its physiological effects in a number of different ways. However, especially in early stage disease, these may be barely discernible. Local effects are due to the physical presence of a tumour, for example polyps in colorectal cancer causing rectal bleeding and anaemia.[12] Systemic effects, for example fatigue and weight loss, are distant effects unrelated to tumour load but are thought to be mediated by tumour necrosis factor-α (TNFα).[13] Metastasis, the spread of cancer to secondary tissues, can produce effects due to tumour load in these secondary tissues, for example a persistent cough due to lung metastases.[14]

Prognosis is variable and much will depend on the disease stage. There are a number of techniques for cancer treatment, including surgery, chemotherapy, radiotherapy, biological or hormonal treatments and transplantation. Factors such as tumour type, tumour location and disease stage influence treatment options. In the majority of cases a combination of therapies are employed.[15]

The origins of chemotherapy can be traced back to the World Wars. The use of chemical warfare in World War I is well documented; autopsies of victims of mustard gas attacks were noted to have leukopenia (a decrease in white blood cells). In World War II, following the air raid on Bari Harbour in 1943 which led to the accidental release of 100 tons of mustard gas,[16] patients were noted to have leukopenia and bone marrow aplasia (cessation of the production of blood cells by the bone marrow). These observations led to the investigation of the therapeutic potential of nitrogen mustard. Marked regression of lymphomas was noted in both animal and human subjects. Following publication of these results after declassification in 1946, mustine (**Figure 1.1**) became the first ever chemotherapy agent.[17]
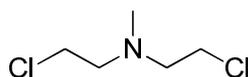
Figure 1.1: Structure of mustine (mechlorethamine)

1.2 Metal based drugs

Several metal ions have key roles in normal biology, for example in signal conduction (sodium and potassium), energy transfer (magnesium), oxygen transport (iron) and electron transfer (copper). Drugs containing metals offer many advantages over traditional organic compounds. Firstly, more diverse coordination numbers and geometries are available compared to carbon based chemistry. Variable oxidation states at the metal centre may allow participation in redox chemistry with the potential to be involved in mechanism of action or drug activation. Compounds may be charged allowing interaction with oppositely charged biomolecules such as negatively charged DNA. Interactions with cellular components may also be facilitated by ligand exchange or aquation processes. Finally, advantage can be taken of properties displayed by metals which are not found in biology.[18] Despite these advantages, the prevalence of metal ions in nature and the historical use of metals in medicine, for example gold-based medicines in Ancient China and Arabia around 1500 BC,[19] research into the medicinal impact of metals was neglected until the latter half of the twentieth century. Interest was renewed with the discovery of the bacteriostatic and bactericidal activity of cobalt and ruthenium complexes by Dwyer in the 1950s.[20] Nowadays, a number of metal based drugs are routinely available for a multitude of diseases including arthritis (gold), syphilis (arsenic), leishmaniasis (antimony) and peptic ulcers (bismuth) among others.[21]

Identification of metal based drugs tends to rely on synthesis of libraries of small molecules followed by screening against cultured cells or in animals. General rules exist to aid in the design of these small molecules, the most well-known being the Lipinski rules.[22] Lipinski's rules state it is more likely that a drug will show poor absorption or permeation if the following criteria are not followed:-

1.   Molecular weight is less than 500 Da
2.   There are no more than 5 hydrogen bond donors
3.   There are no more than 10 hydrogen bond acceptors
4.   The log *P* value (octanol-water partition coefficient) is less than 5

However, these rules are more relevant for organic molecules and thus the two-pole complementary principle was developed for use with inorganic molecules.[23] The criteria for assessment of the anti-cancer activity of metal drugs are as follows:-

1.   Contain hydrophilic and hydrophobic groups
2.   Dipole moment is not equal to zero for active intermediate complex
3.   Appropriate hydrolysis rate under physiological conditions to give an active intermediate with at least two water molecules in the *cis* configuration
4.   Appropriate Pearson "hard-soft" degree
5.   Left handed enantiomer (Λ) if chirality is present

However, it is important that these are used as guides as opposed to strict rules, so as not to restrict the types of complexes researched. For example, a number of *trans* platinum(II) complexes have displayed good anti-cancer activity despite the fact that according to the two-pole complementary principle the *cis* isomer is generally considered preferable.[24, 25] The development of new drugs may take advantage of research that has been carried out for inorganic complexes not in the biological field, for example work on ruthenium Grubb's-type catalysts has helped inform research into the similarly structured ruthenium arene anti-cancer complexes.[26]

The field of medicinal inorganic chemistry changed in 1969 with the publication by Rosenberg of the anti-cancer properties of a previously discovered platinum diammine complex.[27] This paved the way for further research into firstly platinum anti-cancer drugs and then into the anti-cancer potential of a number of metals including antimony, bismuth, cobalt, copper, gallium, gold, iridium, iron, osmium, palladium, rhodium, ruthenium, silver, tin, titanium, vanadium and zirconium. [28-36] Currently, the only approved metal-based anti-cancer drugs are cisplatin, carboplatin, oxaliplatin and arsenic trioxide.

1.3 Cisplatin

Cisplatin is one of the most widely used chemotherapy drugs in the world but was happened upon accidentally in the late 1960s during experiments on the effect of electric currents on cell division in *Escherichia coli* (*E. coli*). [37, 38] *E. coli* cells were grown in ammonium chloride buffer and current was applied using platinum electrodes. As the experiments progressed, the *E. coli* cells were noted to change shape from the classical rod-shape to "long and filamentous" cell structures.[38] This

was found to be due to inhibition of binary fission (the prokaryotic mechanism of cell division) without inhibition of cell growth. This inhibitory factor was a platinum hydrolysis product which formed on the surface of the platinum electrodes. This compound was determined as cis-[PtCl$_2$(NH$_3$)$_2$] (cisplatin) (**Figure 1.2**); the equivalent trans complex was found to be inactive.[38]
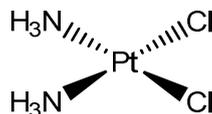
Figure 1.2: Structure of cisplatin

The potential inhibitory effect of cisplatin on eukaryotic cells, specifically cells which had undergone malignant transformation, was probed further by Rosenberg. Cisplatin and its 6-coordinate analogue cis-[PtCl$_4$(NH$_3$)$_2$] were tested on a murine model using transplantable sarcoma 180 tumours. Cisplatin was found to induce complete tumour regression and re-implantation of sarcoma tumours was strongly rejected.[39] Clinical trials were initiated in 1971 and the drug was FDA approved in 1978.[40] Today, almost four decades since approval, cisplatin remains the cornerstone of chemotherapy for bladder, cervical, ovarian, testicular, non-small cell lung cancer and malignant mesothelioma.[41]

The mechanism of action of cisplatin is by induction of apoptosis due to interaction with DNA (**Figure 1.3**). When cisplatin enters the bloodstream, the high chloride environment (approximate chloride concentration 100 mM) prevents exchange of labile chloride ligands with water. Intact cisplatin may be transported into cells passively by diffusion or be actively transported across membranes. Transporters currently associated with this cisplatin active transport are copper transporter 1 (Ctr1), copper transporter 2 (Ctr2), P-type copper transporting ATPases (ATP7A and ATP7B), organic cation transporter 2 (OCT2) and multi-drug extrusion transporter 1 (MATE1).[42] The low intracellular chloride environment (approximate chloride concentration 10 mM) allows exchange of one or both chloride ligands with water to form a charged complex.
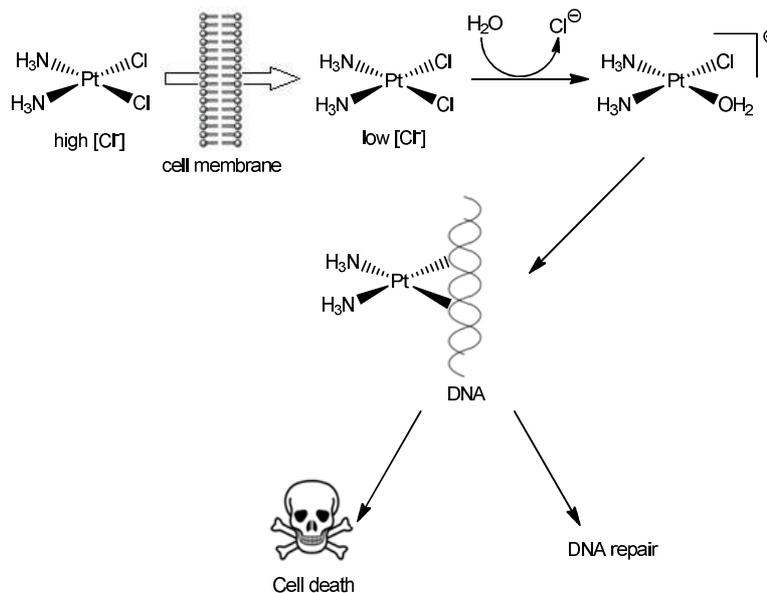
Figure 1.3: Mechanism of cisplatin action[43]

These charged platinum species can enter the nucleus and interact with electron dense regions of DNA. The primary site of interaction is the N7 atom of guanine; however, interaction with lone pair nitrogen atoms on adenine and cytosine also occurs, resulting in the formation of a DNA adduct (**Figure 1.4a**). Interaction of cisplatin with an additional residue results in ring closure and the formation of the bifunctional adduct. Adducts formed may be either interstrand or intrastrand and may consist of either 1,2 or 1,3-crosslinks; the most common adducts formed are intrastrand Pt-GG (47 - 50 %) or Pt-AG (23 - 28 %).[44-48] Crosslink formation leads to destacking of purine and pyrimidine rings and thus introduces a 32 - 35° bend towards the major groove into the helix[49] (**Figure 1.4b**), leading to either DNA repair *via* nucleotide excision repair or initiation of apoptosis.[50, 51]

Cisplatin induced DNA damage activates numerous factors including p53, c-Abl, caspases 6 and 7, PKC, ERK and various microRNAs causing cell cycle arrest. Downstream signalling and activation processes lead to amplification of pro-apoptotic factors and imbalance against anti-apoptotic factors within the cell leading to apoptosis.[52]
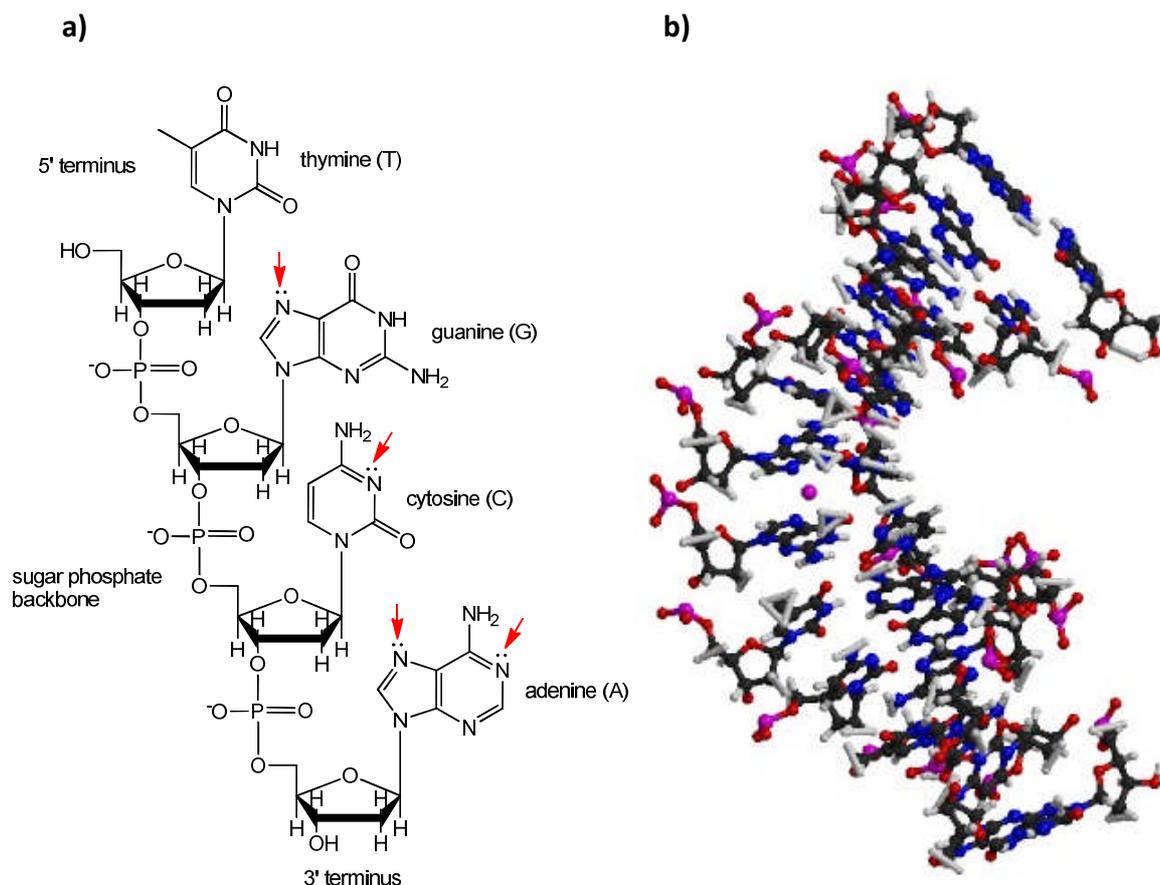
Figure 1.4: Cisplatin binding to DNA a) DNA binding sites for cisplatin[53] b) cisplatin-DNA adduct (intrastrand Pt-GG) showing helical distortion[54]

Cisplatin is a successful drug with impressive potency e.g. 90 % cure rate for testicular cancer.[55] However, there are a number of issues and side-effects associated with cisplatin therapy which limits its clinical use. One problem with cisplatin is that following intravenous administration, cisplatin can interact with a number of plasma proteins, particularly those containing thiol groups.[56] Thiol containing proteins are abundant both extracellularly and intracellularly, for example albumin and glutathione respectively. This limits the bioavailability of cisplatin thus reducing the elimination half-life and limiting efficacy; it may also cause drug deactivation[57] and adverse drug reactions (ADRs).[58] Another of the main problems with cisplatin are the severe ADRs which can affect patient compliance and carry an associated inherent risk of morbidity and mortality. The most common and clinically important ADRs include nephrotoxicity, neurotoxicity, ototoxicity and emesis. Nephrotoxicity, occurring in approximately a third of patients, results from direct injury to the renal tubules leading to a reduction in glomerular filtration rate and electrolyte disturbances; severe damage can result in acute kidney injury.[59, 60] Neurotoxicity is characterised by a sensory neuropathy leading to neuropathic pain and paresthesias; the mechanism involved is unclear but may involve mitochondrial

changes and activation of protein kinases.[61] Ototoxicity is a significant problem; especially in paediatric patients where 60 % will suffer permanent bilateral hearing loss, which in turn may affect speech and language development with subsequent social and educational consequences.[62] A further problem is the development of resistance, which may force increased dosage with the subsequent increase in risk or severity of ADRs and of course eventual discontinuation of cisplatin therapy. Intrinsic and acquired resistance is seen in a number of cancers e.g. colon cancer (intrinsic), ovarian and non-small cell lung cancer (acquired). The mechanisms involved in cisplatin resistance include reduction in cellular uptake mediated by transporters, production of intracellular thiol-containing proteins and increased capacity for DNA repair and tolerance of DNA damage.[63-65]

1.4 Alternative platinum anti-cancer drugs

Cisplatin paved the way for research into metal-based anti-cancer drugs; but despite its huge success, problems such as toxicity, resistance, the intravenous administration route and inactivity against common cancers such as lung cancer have encouraged research into new platinum anti-cancer drugs. To date, only two other platinum anti-cancer drugs have gained FDA approval: carboplatin in 1989 and oxaliplatin in 2002 (**Figure 1.5**).[53] Both carboplatin and oxaliplatin are structurally similar to cisplatin with a square planar structure around a central platinum(II) atom with two amine groups in the *cis* configuration, and thus these drugs exert their anti-cancer effects *via* a similar pathway.
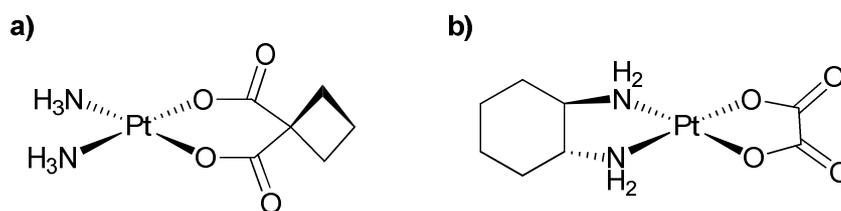
Figure 1.5: Structures of a) carboplatin b) oxaliplatin

In carboplatin, the leaving group is a cyclobutanedicarboxylate which is more stable than the chloride leaving groups in cisplatin. The aquation of carboplatin is thus slower than cisplatin, which has led to the suggestion that the mechanism of action differs from cisplatin which requires aquation for activity.[66] This decreased hydrolysis rate also means that carboplatin shows vastly reduced binding to thiol-containing proteins compared to cisplatin and hence greater bioavailability and a longer

half-life.[37, 67] The main advantage of carboplatin over cisplatin is the reduced toxicity, with vastly reduced nephrotoxicity and less severe emesis;[68] however a greater degree of myelosuppression is seen.[69] Carboplatin is FDA approved for ovarian and non-small cell lung cancer[70] where it has replaced cisplatin in the majority of cases.

Oxaliplatin (**Figure 1.5b**) is FDA approved for colorectal and stage III colon cancer and is being investigated for other cancers.[71] It is currently the only platinum-based drug to show activity against colorectal cancer, especially when used in combination with 5-fluorouracil and leucovorin.[72] Interestingly, oxaliplatin displays some activity against cisplatin resistant cell lines, implying a different mechanism of action.[73] Oxaliplatin contains an oxalate leaving group and a diaminocyclohexane ligand, the *R,R* isomer of this non-leaving group ligand is more effective than the *S,S* isomer due to the ability to form hydrogen bonds between the ligand amine and the guanine oxygen in a Pt-GG lesion.[74, 75]

In addition to carboplatin and oxaliplatin, there are three platinum anti-cancer drugs, heptaplatin, lobaplatin and nedaplatin (**Figure 1.6**), which are licensed in individual countries: Korea, China and Japan respectively.[76] A number of platinum drugs are also under investigation in either pre-clinical or clinical trials. Areas of research include adaptations of cisplatin to improve tissue and cellular targeting such as the addition of glucose or amino acid residues, investigation into novel platinum(II) drugs such as polynuclear platinum drugs or drugs based on transplatin, investigation into platinum(IV) pro-drugs for the release of cisplatin e.g. satraplatin, iproplatin and improved delivery of cisplatin through the use of nanoparticles, carbon nanotubes and polymeric micelles e.g. ProLindac, lipoplatin.[77]
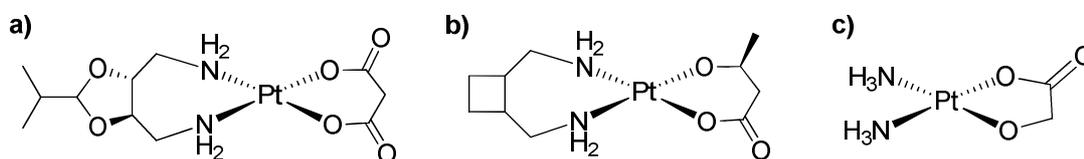
Figure 1.6: Structures of a) heptaplatin b) lobaplatin c) nedaplatin

1.5 Ruthenium anti-cancer drugs

After platinum, ruthenium is the metal undergoing the most extensive investigation in anti-cancer research, with three ruthenium-based drugs having undergone clinical trials: NAMI-A, KP1019 and NKP1339. Ruthenium has a number of useful properties for drug design: it can exchange with *O* and *N* donors in a similar way to platinum and with comparable kinetics allowing complexes to reach their target without becoming fully dissociated. Under physiological conditions three oxidation states (II, III, IV) are accessible and thus ruthenium(III) or ruthenium(IV) prodrugs can be reduced to the more active ruthenium(II) in the hypoxic tumour environment, allowing improved tumour selectivity. Ruthenium can also mimic iron to be transported into tumour cells *via* transferrin receptors, again allowing greater selectivity.[78]

One of the first ruthenium complexes to be investigated was the cytological dye "ruthenium red" which inhibits mitochondrial calcium transport through interaction with membrane transporters.[79] Other early compounds included six coordinate ruthenium analogues of cisplatin.[80] The major limiting factor against the utility of these complexes was poor aqueous solubility, hence the highly soluble ruthenium(II)-DMSO complexes were investigated.

1.5.1 NAMI-A

Imidazolium [*trans*-imidazole-DMSO-tetrachlororuthenate(III)] (NAMI-A) (**Figure 1.7a**) was the first ruthenium anti-cancer drug to enter clinical trials. This drug was initially synthesised as the sodium salt (NAMI) (**Figure 1.7b**), however this led to co-precipitation of DMSO with the sodium ion and therefore the sodium counterion was replaced with an imidazolium ion to give NAMI-A.[81] NAMI-A is stable in the solid state but in solution undergoes pH dependent aquation of two chloride ligands followed by the DMSO ligand leading to the formation of a polyoxo species.[82-84]
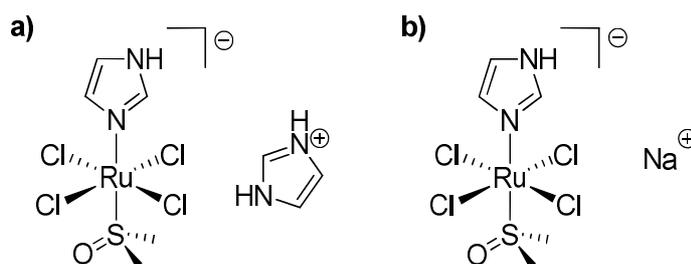
Figure 1.7: Structure of a) NAMI-A b) NAMI

NAMI-A displayed potent anti-metastatic activity against lung metastases of several solid tumours, especially non-small cell lung cancer (NSCLC), as opposed to activity against the primary tumour. This is important as metastasis is the major cause of cancer mortality and the lungs are the primary site of metastasis.[14, 83] In addition, this anti-metastatic activity was coupled with low systemic toxicity. Initial clinical trial results were promising with considerable reduction in metastasis and improved survival times.[85-87] However, NAMI-A did not progress beyond phase II clinical trials due to insufficient efficacy in NSCLC patients when co-administered with gemcitabine (a licensed NSCLC drug) compared to gemcitabine alone.[88] Interestingly, NAMI-A has also been shown to have anti-plasmodial activity against *Plasmodium falciparum*, the major parasite of four species responsible for malaria.[89]

Despite extensive research, the mechanism of action of NAMI-A is uncertain. NAMI-A can transiently block the cell cycle at G2M phase[90] and bind to both DNA and RNA,[91, 92] however these interactions are observed at higher concentrations and are not thought to be physiologically relevant. It is more likely that the mechanism of action involves inhibition of angiogenesis through inhibition of vascular endothelial growth factor (VEGF),[93] cytoskeletal remodelling through the disruption of matrix metalloproteinase (MMP) release[94] and interaction with actins and collagens,[87, 95, 96] and facilitation of the interaction between cancer cells and tumour infiltrating lymphocytes among others.[97]

1.5.2 KP1019 and NKP1339

Indazolium [*trans*-tetrachloro-*bis*(indazole)-ruthenate(III)] (KP1019) (**Figure 1.8a**) was developed from the analogous imidazole complex (KP418) (**Figure 1.8b**) which displayed high activity against rat colorectal carcinoma models.[98] KP1019 shows improved activity in the rat colorectal cancer model than KP418 with reduced mortality and weight loss; it was also found to be more active than 5-fluorouracil (a licensed colorectal cancer drug).[99] NKP1339 (**Figure 1.8c**) is the sodium salt of KP1019 and displays improved solubility and formulation characteristics and is the current lead compound for this class of drugs.[100, 101]
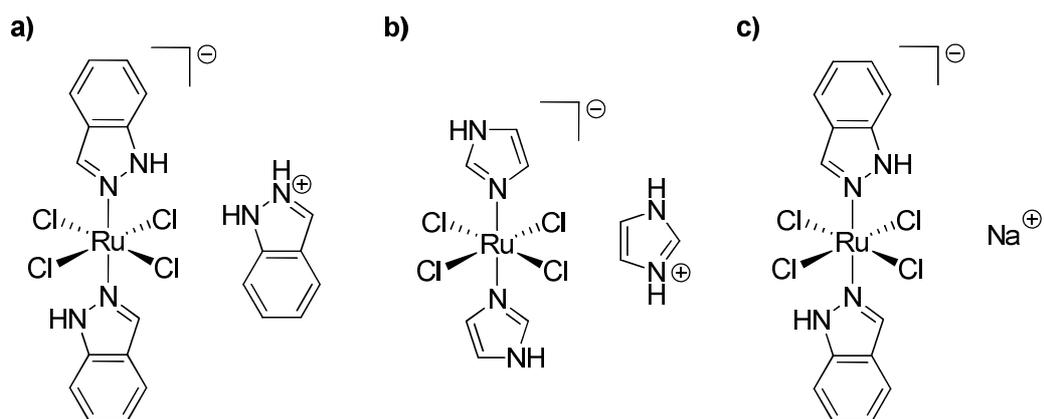
Figure 1.8: Structures of a) KP1019 b) KP418 c) NKP1339

KP1019 displayed promising activity with low systemic toxicity in phase I trials for patients with a variety of advanced solid tumours. The clinical effects did not appear to correlate with the dosage. Pharmacokinetic analysis found that the drug was bound mainly to plasma proteins with a long half-life and low clearance.[102] Due to solubility issues, further clinical trials were performed with NKP1339. Anti-cancer activity was promising with NKP1339, but less potent compared to KP1019, with low systemic toxicity. It is therefore thought that these two drugs share similar mechanisms of action.[103, 104]

KP1019 and NKP1339 are thought to undergo reduction from ruthenium(III) to ruthenium(II) following rapid intracellular uptake,[103] as opposed to when bound to plasma proteins such as albumin and transferrin.[105, 106] The aquation of the drugs is accelerated upon reduction to ruthenium(II) resulting in a species which shows greater reactivity with biomolecules.[107-109] KP1019 and NKP1339 have been found to induce the formation of reactive oxygen species (ROS)[110, 111] which can then lead to damage to the mitochondria. In addition, NKP1339 is a direct nitric oxide scavenger which suggests that this drug may induce effects in cell migration and angiogenesis.[93]

1.5.3 Ruthenium arene complexes

The most numerous group of ruthenium anti-cancer compounds currently under investigation are the ruthenium arene complexes. Ruthenium arene complexes tend to consist of ruthenium(II) stabilised by a facially bound η^6-arene, often one or two labile ligands are present. The two main groups of ruthenium arene compounds which have been studied are RAPTA complexes and RAED-type complexes (**Figure 1.9**).
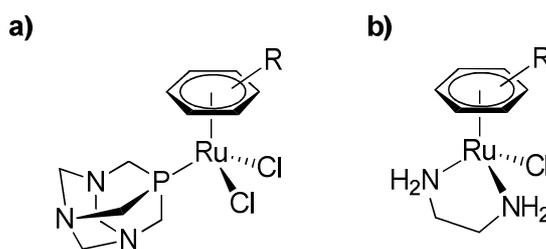
Figure 1.9: General structures of a) RAPTA b) RAED type complexes

RAPTA complexes are "piano stool" complexes developed by Dyson and contain a η^6-arene, an amphiphilic monodentate 1,3,5-triaza-7-phosphatricyclodecane (pta) ligand and two free coordination sites which tend to be occupied by labile chloride ligands. The pta ligand provides aqueous solubility dependent upon the nature of the arene ligand, more electron withdrawing groups leading to poorer stability, which allows improved hydrolytic stability and facilitates drug administration.[112, 113]

The first RAPTA complex, RAPTA-C (**Figure 1.10a**), displayed *in vitro* DNA damaging ability in a pH dependent manner and therefore it was hypothesised that these complexes could provide selective DNA damage in hypoxic tissue such as cancer cells.[114] A large number of different RAPTA complexes with variations to the arene ring and substituents of the pta ligand have been investigated for their anti-cancer potential. In general, these complexes displayed moderate activity towards TS/A adenocarcinoma cell lines with no toxicity towards the non-cancerous HBL-100 cell line. RAPTA-T (**Figure 1.10b**) in particular displayed good selectivity for the cancerous cells.[115] RAPTA-T also displayed potent anti-metastatic activity and appeared selective for invasive cancer cells and metastases. This activity is thought to be due to the action of RAPTA-T on cell surface molecules and the cytoskeleton leading to the loss of cellular flexibility which is required for cellular detachment and tissue invasion.[116] Replacement of the chloride ligands with more stable leaving groups has little effect on activity and thus hydrophobicity can be tuned by the exchange of chloride ligands for neutral two electron donor ligands such as imidazole.[113] Both RAPTA-C and RAPTA-T are able to interact with DNA and a number of different proteins e.g. chromatin,[117] glutathione,[118] cytochrome c,[119] superoxide dismutase[120] and metallothioneins,[121] and inhibit different enzymes e.g. thioredoxin reductase, cathepsin B[122] and PARPs;[123] these interactions are thought to be important for the mechanism of drug action which has yet to be elucidated.
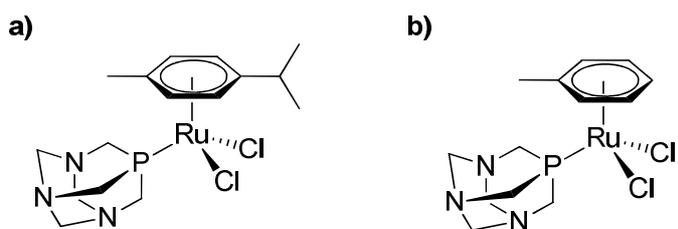
Figure 1.10: Structures of a) RAPTA-C b) RAPTA-T

RAED-type complexes were initially investigated by Sheldrick who demonstrated the anti-tumour potential of a ruthenium(II) ethylenediamine chloride hexafluorophosphate salt (**Figure 1.11**).[124] Extensive development of this original structure has been carried out by Sadler, with variation of both the arene and bidentate ligand.[125] In these ruthenium arene complexes, fine tuning of the chelating ligand can improve aqueous stability and control ligand exchange kinetics; the arene can influence cellular transport and the leaving group can assist in controlling the activation.[126] Structure-activity relationship investigations have revealed that the optimum structures contain a stable bidentate *N,N* donor ligand, a hydrophobic arene and a labile halide ligand.[124, 125, 127]
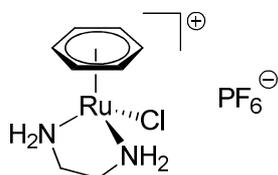
Figure 1.11: Structure of $[(\eta^6\text{-benzene})Ru(en)Cl]PF_6$

The anti-cancer potential of a wide variety of these ruthenium arene complexes has been studied; cytotoxicity was noted against ovarian cancer and NSCLC cell lines, with an increase in activity correlating with increase in arene ring size (**Figure 1.12**), a trend which is also noted for hydrolysis and hydrophobicity with ring size.[126-128]
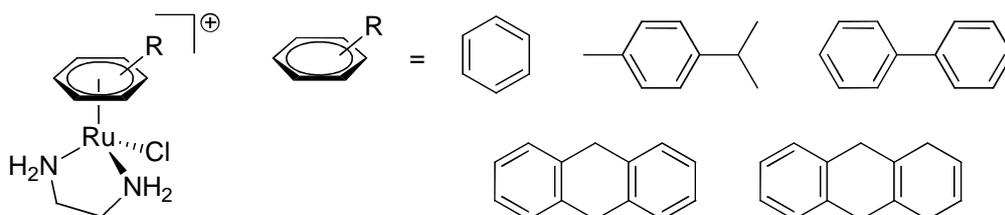
Figure 1.12: Ruthenium arene complexes with increasing arene ring sizes

Variation of the ligand in ruthenium *p*-cymeme complexes also has significant implications for activity (**Figure 1.13**). Complexes containing two monodentate *N*-donor ligands as opposed to the bidentate ethylenediamine (en) ligand show very low activities.[125] Bidentate *N,N* and *N,O* ligands based on amino acids were less cytotoxic than the en ligand. Replacement of the σ-donor en ligand with π-acceptor 2,2'-bipyridine ligand led to almost complete inactivation.[129] Amongst polar substituents to the bidentate ligand, 2-phenoxy derivatives display the most potent activity but generally non-polar substituents conferred greater activity than polar substituents.[129] In order to improve DNA capacity, the neutral en has been replaced with an anionic β-diketonate ligand. These complexes can hydrolyse readily and display selective binding to adenosine.[130]
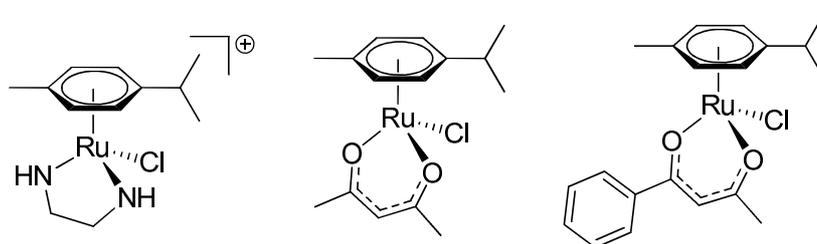
Figure 1.13: Ruthenium *p*-cymene complexes with varying bidentate ligands

In general, it appears that ruthenium arene complexes with labile halide ligands undergo aquation intracellularly. Similar to cisplatin, the rate of aquation can vary substantially depending upon the nature of the bidentate ligand;[131] for example acetylacetonate (acac) containing complexes aquate more rapidly than en containing complexes. This is ascribed to the more electron donating nature of the acac ligand.[130, 132] Interaction with DNA is thought to be crucial to the mechanism of action as ruthenium(II) can bind to guanine and thymidine residues.[132, 133] The preference for guanine binding may allow these drugs to target guanine rich regions of DNA such as the telomeres (repeat sequences which act as caps at the ends of chromosomes). A ruthenium arene complex with an en ligand has the ability to unwind supercoiled DNA and induce interstrand and intrastrand Ru-GG crosslinks similar to cisplatin. Overall, fewer interactions with proteins are observed which may account for the low systemic toxicity and aid the transport of drugs to the target cells.[134]

1.6 Rhodium and iridium anti-cancer drugs

In contrast to ruthenium, there have been relatively few investigations into the biological activity of the neighbouring metal rhodium. The discovery that dimeric rhodium(II) carboxylates (**Figure 1.14**) exhibit anti-cancer activity prompted increased research into rhodium complexes.[135-138] These type of complexes display improved cytotoxicity correlating with greater lipophilicity which in turn is dependent on the size of the alkyl group attached to the carboxylate.[139] The mechanism of action is unclear; it may be through DNA binding akin to cisplatin or inhibition of enzymes involved in DNA synthesis due to complex binding to protein sulphydryl sites.[140, 141]
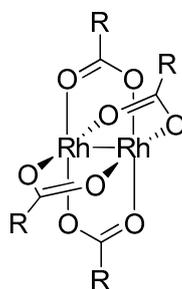
Figure 1.14: General structure of rhodium carboxylates complexes

Recently, rhodium carboxylate complexes containing bipyridine or phenathroline ligands have displayed cytotoxicity.[142, 143] A range of carboxylate complexes have also been shown to increase the radiation sensitisation of cancer cells *in vitro*, an effect ascribed to their ability to deplete intracellular thiols.[144] Similar complexes with various anti-malarial drugs as additional ligands also show activity, with the most active complex containing the drug mepacrine.[145]

The most widespread investigation into rhodium drugs has been through the synthesis of rhodium analogues of previously known ruthenium(III) complexes, these tend to show lower activity as they are unlikely to be activated by reduction unlike their ruthenium counterparts.[146, 147] However, some complexes defy this trend, for example a rhodium chloride complex with diaminopyridine displayed significant anti-tumour effects while in addition displaying no greater nephrotoxicity than cisplatin.[148]

There have been several studies on rhodium complexes with polyaromatic ligands such as modified bipyridyl, phenylpyridyl or phenathroline ligands, inspired by previously known rhodium complexes with luminescent properties. The large planar aromatic nature of these ligands is attractive as there is the potential for DNA intercalation. Complexes were found to be cytotoxic against a number of cell lines with the polyaromatic ligands providing increased hydrophobicity and altering the metal redox properties. However, despite the intercalating ability of the aromatic ligands DNA did not appear to be the primary target.[149] The nature of the polyaromatic ligand had a significant effect on cytotoxicity; for example in facial rhodium(III) polypyridyl complexes (**Figure 1.15**) potency was increased in the order dppn > dppz > dpq, phen >> bpy.[150-153] Other similar complexes display anti-cancer effects through a number of different mechanisms including inhibition of various enzymes,[154, 155] inhibitors of protein-protein interactions,[156] oxidative DNA damage[157] and through DNA intercalation.[158] One intercalator in particular shows specific binding to destabilised regions in DNA close to base pair mismatches.[159]
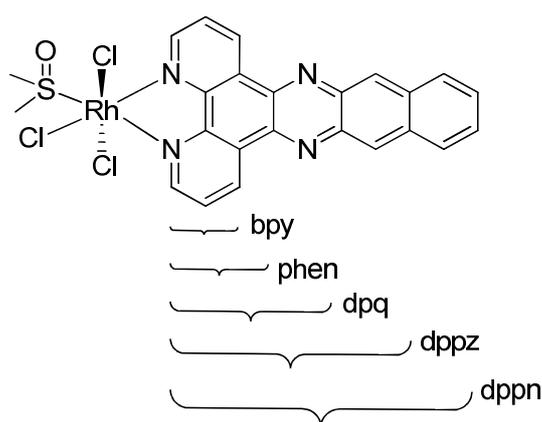
Figure 1.15: Structures of Rh(III) *fac*-[RhCl$_3$(DMSO)] with 2-2' bipyridine (bpy), 1,10-phenathroline (phen), dipyrido[3,2-d:2',3'-f]quinoxaline (dpq), dipyrido[3,2-a:2',3'-c]phenazine (dppz) and benzo[*i*]dipyrido[3,2-a:2',3'-c]phenazine dppn ligands

A variety of rhodium(I) complexes have also displayed anti-cancer activity including square planar *cis*-1,5-cyclooactadiene (COD) complexes with a number of additional ligands such as acacs.[160, 161] Some of these complexes displayed anti-metastatic effects which are thought to be due to modifications occurring at the primary tumour site.[162] Cationic rhodium COD complexes containing anti-parasitic drugs also show cytotoxicity,[163] as do rhodium carbonyl complexes with sulphonamide derivatives which are cytotoxic to tumour cells without associated nephrotoxicity.[164]

As with rhodium, there has been relatively little investigation into the anti-cancer potential of its fellow group IX metal iridium. Iridium complexes have frequently been studied as analogous complexes for known ruthenium(III) complexes. Such iridium complexes, similar to the analogous rhodium complexes, tend to show lower cytotoxicity due to the reduced likelihood of activation by reduction or by ligand exchange/aquation due to less rapid exchange kinetics.[165] However, this relative inertness to substitution can be exploited to form kinetically inert complexes which can act as enzyme inhibitors. Iridium complexes with pyridocarbazole act as a structural mimics of staurosporine, a protein kinase inhibitor (**Figure 1.16**).[166] This type of complex selectively inhibits Flt4 kinase, a protein kinase involved in the maintenance of the lymphatic system, and shows good cytotoxicity results. Variations in the axial ligands can result in selectivity for different protein kinases.[167]
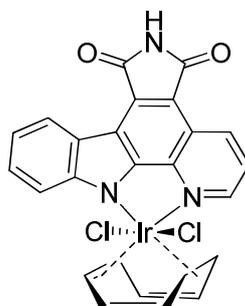
Figure 1.16: Iridium(III) pyridocarbazole complex

Several iridium arene "piano stool" complexes have been investigated by Sadler as counterparts to the ruthenium arene complexes (**Figure 1.17**). The iridium(III) centre is stabilised by pentamethylcyclopentadiene (Cp*), which also enhances the lability of a monodentate ligand which helps overcome issues associated with slow ligand exchange reactions as for the analogous ruthenium complexes ligand exchange appears to be an important step in the mechanism of action.[168]
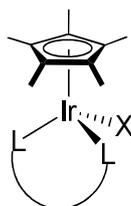
Figure 1.17: General structure of iridium arene complexes

Cytotoxicity of these complexes was found to be dependent upon the nature of the arene, complexes with phenyl or biphenyl substituted Cp* as the arene were more active than complexes containing Cp* as the arene moiety. Enhanced cytotoxicity was correlated with increased hydrophobicity, cellular accumulation and DNA binding and intercalating ability.[31] The nature of the bidentate ligand also has a large effect on cytotoxicity with complexes containing phenylpyridine ligands (*C,N* coordination) showing enhanced cytotoxicities compared to the analogous complexes containing bipyridine ligands (*N,N* coordination).[169] Complexes containing ketoiminate ligands (*N,O* coordination) were also found to have improved activity compared to complexes containing picolinamide (*N,N* coordination) or napthoquinone ligands (*O,O* coordination).[170, 171]

Iridium complexes with polyaromatic ligands, similar to the previously discussed rhodium complexes, have also been investigated. Similar trends to the rhodium complexes are noted with improved cytotoxicity with an increase in the length of the polyaromatic ligand, despite the fact that DNA interaction or intercalation is not observed at a significant level.[150, 151, 157] Facial isomers were found to be more toxic than the meridional isomers, in contrast to the rhodium complexes.[172] Further studies with the 5,6-dimethylsubstituted phenathroline ligand showed that these complexes displayed remarkably improved activity compared to the unsubstituted phenathroline analogues[158] and that the mechanism of action was through the production of ROS leading to apoptosis through the intrinsic mitochondrial pathway.[152]

1.7 Cobalt anti-cancer drugs

Research into cobalt containing drugs is in its infancy in comparison with metals such as platinum and ruthenium. However, cobalt has a number of useful properties which may be exploited in drug design: under physiological conditions two oxidation states (II, III) are accessible and thus cobalt(III) prodrugs can be reduced to more active cobalt(II) drugs in a hypoxic tumour environment; this allows improved tumour selectivity.[173] Cobalt(III) is generally kinetically inert; however it may undergo ligand exchange reactions, such as aquation, with comparable kinetics to platinum(II) and ruthenium(III) which allows the potential for the activation by aquation mechanism.[165] Cobalt is an essential trace element and as such is relatively non-toxic; this allows the creation of potential drugs with less systemic toxicity than is noted with for example platinum based drugs. In addition, as cobalt is present

physiologically there are transport mechanisms for the movement of cobalt containing molecules such as cobalamin into cells which may be exploited for improved drug delivery.

The first investigations into the biological potential of a variety of cobalt(III) complexes were described by Dwyer in 1952 (**Figure 1.18**). These complexes were found to induce death in mice through paralysis and respiratory failure. In addition, the 1:10-*bis*(salicylideneamino)-4,7-dithiadecane cobalt iodide complex (**Figure 1.18e**) exhibited anti-bacterial properties against both *Escherichia coli* and *Staphylococcus haemolyticus*.[20] Further investigations into charged and neutral cobalt complexes found these complexes to be potent competitive inhibitors of cholinesterase, hypothesised to be the mechanism of action.[174] This work led to the investigation of substituted phenathroline cobalt complexes which showed potent anti-bacterial activity.[175] The anti-cancer potential of these complexes were not investigated and despite the promising anti-bacterial properties, the potential of cobalt complexes as drugs was overlooked for many years.
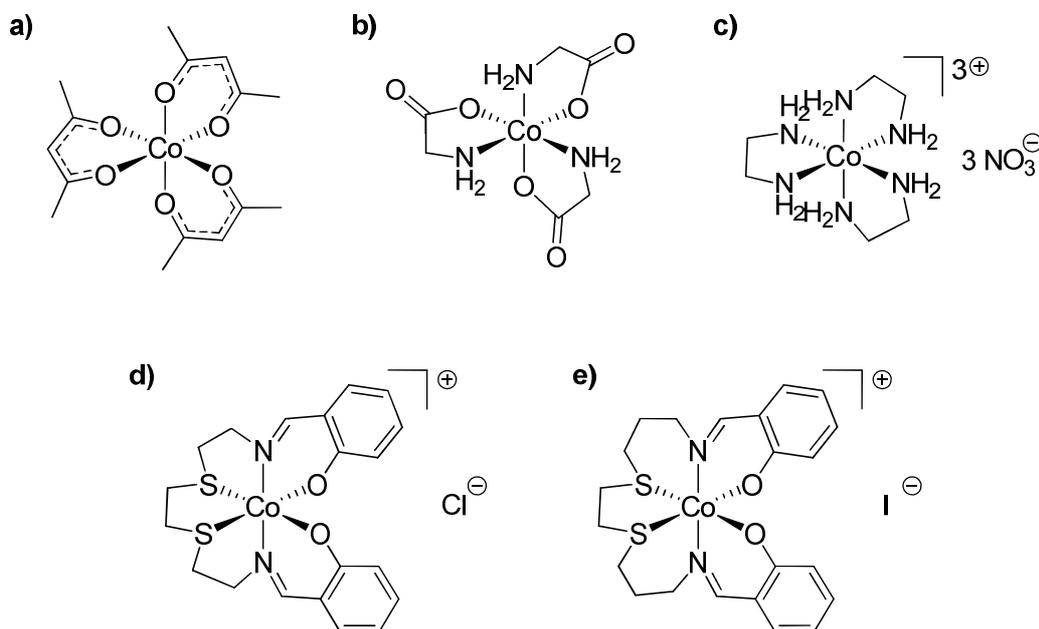
Figure 1.18: Cobalt complexes investigated by Dwyer a) cobalt *tris*-acetylacetone b) cobalt *tris*-glycine c) cobalt *tris*-ethylenediamine nitrate d) cobalt 1:8-*bis*(salicylideneamino)-3,6-dithiaoctane chloride e) cobalt 1:10-*bis*(salicylideneamino)-4,7-dithiadecane iodide

1.7.1 Cobalt complexes with non-bioactive ligands

There are currently no FDA approved cobalt-containing drugs, however Doxovir (CTC-96) is currently in Phase II clinical trials as an anti-viral agent. Doxovir is a cobalt(III) Schiff base complex with two axially bound imidazole derivatives (**Figure 1.19**) which exhibits potent activity against drug resistant strains of herpes simplex virus 1 (HSV-1) and has shown to be effective in the treatment of herpetic keratitis and adenovirus keratoconjunctivitis.[176-179] The mechanism of action is thought to involve exchange of an axial ligand for a histidine residue at the active site of a HSV-1 serine protease.[180, 181] Variation in the axial ligand permits binding to histidine residues of zinc finger motifs, common to many transcription factors, preventing interaction of the transcription factor with its specific response element in DNA. This has been shown to interrupt transcription and cell signalling pathways in cancer cell lines.[180, 182-186] Other histidine containing enzymes e.g. thermolysin, α-thrombin and MMP-2 may also be targeted by Doxovir.[181, 187]
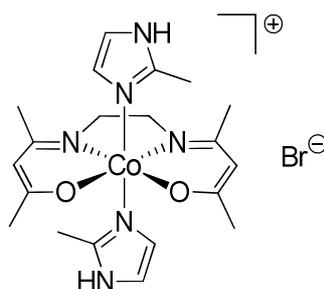
Figure 1.19: Structure of Doxovir (CTC-96)

Following the discovery of Doxovir and its ability to disrupt signalling pathways involved in cancer, a few isolated investigations into the anti-cancer potential of various cobalt(II) and cobalt(III) Schiff base complexes have been performed. Cobalt complexes based on the structure of Doxovir, with the same Schiff base backbone and variable axial ligands including nicotinamide and isonicotinamide, were found to undergo aquation readily under physiological conditions and exhibited potent anti-tumour and anti-metastatic activity in a mouse model. The mechanism of action is thought to be through the interactions with MMPs and the induction of ROS leading to DNA damage and apoptosis.[188] The biological potential of cobalt(II) and cobalt(III) Schiff base complexes containing hydroxyaminoethylamine, phenylmethylamine, ethylenediamine, hydroxybenzylideneamine and aminophenyl morpholine derivatives has been investigated. The lack of understanding of structure-activity relationships in cobalt Schiff base complexes is apparent when examining these studies as the complexes with ethylenediamine and hydroxybenzylideneamine

moieties showed moderate activity, whereas complexes with morpholine derivatives were non-toxic; ethylamine and phenylmethylamine derivatives were found to inhibit urease, but their anti-cancer potential was not evaluated.[189-192] Schiff base complexes with heterocyclic substituents showed moderate activity against cancer cell lines with reduced toxicity to non-cancerous cell lines and a tridentate Schiff base complex containing a phenylimiomethyl derivatives was non-toxic.[193, 194] A cobalt(II) Schiff base complexes with isonicotinic hydrazine moieties formed a water soluble coordination polymer showed good DNA and protein binding, radical scavenging and good anti-cancer activity with low systemic toxicity.[195]

Coordination complexes of cobalt with other non-Schiff base type ligands have also been investigated. Cytotoxicities in the low micromolar range were observed in cobalt complexes with macrocyclic ligands[196] in comparison to a moderately active cobalt thiocarbazate complex[197] and inactive cobalt phenathroline maltol complexes.[198] In contrast, other cobalt phenathroline complexes had promising anti-cancer activities with some selectivity for cancerous cells over non-cancerous cells and evidence for DNA cleavage and inhibition of topoisomerase I.[199-203] When surfactant groups were included activity was often improved with enhanced DNA damage and induction of apoptosis.[204-206] Cobalt thiosemicarbazone complexes have shown good to moderate anti-cancer activities with the ability to inhibit DNA synthesis and induce DNA strand breaks.[207-210] A heterobimetallic ruthenium-cobalt complex with phenathroline ligands has been shown to undergo photoreactive release of the cobalt centre following irradiation with visible light and reduction of the cobalt(III) centre to cobalt(II).[211] A cobalt complex with bipyridine and azide ligands (**Figure 1.20**) showed good anti-cancer activity with strong minor groove DNA binding, plasma protein binding and the ability to cleave plasmid DNA by the oxidative pathway *via* the induction of hydroxyl radicals.[212]
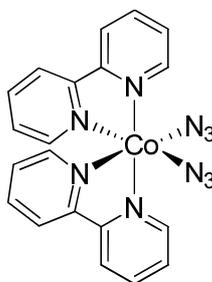
Figure 1.20: Cobalt bipyridine-azide complex

A cobalt polyquinoline complex with good anti-cancer activity has been found to intercalate DNA and induce cleavage of plasmid DNA.[213] A cobalt flumequine (a fluoroquinolone antibiotic) complex was shown to intercalate DNA but was non-toxic to cancer cells, unlike the analogous copper and zinc complexes which were cytotoxic.[214] Organometallic cobalt carborane and supramolecular ruthenium-cobalt heterobimetallic rectangles have also shown promising anti-cancer properties, with the rectangles able to modulate autophagy and induce apoptosis.[215, 216]

1.7.2 Cobalt complexes with bioactive ligands

The dicobalt(0) hexacarbonyl cluster, $[Co_2(CO)_6]$, attached *via* an alkyne linker is known to modify the activity of small molecules.[217] These species may more readily form carbenium ions adjacent to the cobalt carbonyl cluster, which allows the possibility of targeting areas of negative charge such as the sugar phosphate backbone of DNA.[218] A variety of cobalt carbonyl acetylene complexes were investigated for their anti-cancer activity with the lead complex being identified as Co-ASS, a cobalt carbonyl complex with an aspirin derivative (**Figure 1.21**).[219-222] Studies on further cobalt aspirin derivatives found that the hydrophobic aspirin moiety aided cell uptake, presumably through passive diffusion; however hydrophobicity and cell uptake did not determine the cytotoxicity.[220] Aspirin is a non-steroidal anti-inflammatory drug (NSAID) with moderate anti-cancer activity through the inhibition of cyclooxygenase (COX) enzymes leading to cell cycle arrest and apoptosis.[223] Co-ASS was found to be a potent inhibitor of both COX-1 and COX-2 which correlated with cytotoxicity.[224] The differences in the properties of aspirin and Co-ASS may be explained by differences in mechanism as aspirin causes acetylation of serine residues at the active site of the COX enzymes whereas Co-ASS causes acetylation of lysine residues in the substrate channel of the COX enzymes. In addition, Co-ASS showed pro-apoptotic, anti-metastatic and anti-angiogenic properties mediated by the inhibition of NF-κB and MMPs and induction of caspases, implying that the mechanism of action is in fact multifactorial.[224] Co-ASS has also been observed to reduce intracellular ROS suggesting the intracellular release of carbon monoxide (CO), i.e. Co-ASS acts as a CO-releasing molecule (CORM).[225] Introduction of a cyclopentadienyl (Cp) component to coordinate the cobalt carbonyl cluster led to improved cytotoxicity and COX inhibition in comparison to aspirin but not in comparison to Co-ASS.[226]
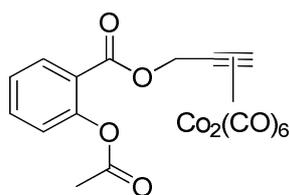
Figure 1.21: Structure of Co-ASS

Coordination complexes of cobalt with other NSAIDs including mefenamic acid,[227-230] tolfenamic acid,[231, 232] naproxen,[233, 234] meloxicam,[235, 236] diclofenac,[237] diflunisal, flufenamic acid and niflumic acid[230] have also been prepared. These complexes show several promising biological functions such as strong DNA and plasma protein binding, high anti-oxidant capacity and inhibition of carbonic anhydrase. However, anti-cancer activities have only been studied for a few of these complexes with promising results against cancer cell lines and a cancer stem cell line.[228, 233] Organometallic cobalt complexes of tamoxifen derivatives (a breast cancer drug), with a cobalt arene moiety replacing a phenyl ring of the parent drug, showed strong oestrogenic activity with the cobalt hydroxytamoxifen derivatives and good cytotoxicities for the cobalt *bis*-aminoethoxytamoxifen derivatives.[238]

A few examples of dicobalt hexacarbonyl complexes with small molecules other than NSAIDs have also been investigated. Complexes with deoxyuridine and fructopyranose derivatives showed only moderate anti-cancer activity.[239, 240] Complexes with imidazoline ligands as oestrogen carriers showed good oestrogenic activity but poor cytotoxicity.[241] Complexes with targeting peptides such as encephalin and neurotensin have also shown good to moderate anti-cancer activity.[242, 243]

Some dicobalt hexacarbonyl complexes have been shown to act as CORMs, similar to one of the Co-ASS derivatives investigated. Initial studies on the $[Co_2(CO)_6]$ cluster with simple alkyl and aryl ligands attached *via* an alkyne found that the ligand had a large effect on the number of CO molecules released, the rate of CO release, cytotoxicity and cell viability.[244] Further investigations with more diverse ligands has found that the mechanism of action may be through cell cycle inhibition and induction of ROS generation leading to apoptosis by the loss of mitochondrial membrane potential and initiation of the late apoptotic pathway.[245, 246]

1.7.3 Bioreductive cobalt prodrugs

A prodrug is an inactive compound which can be activated or metabolised to a form a drug.[247] Cobalt has two readily available oxidation states (II and III) with significant differences in lability and reactivity; conversion between the two states may be synonymous with the conversion from prodrug to drug. The local microenvironment of a tumour is often deprived of oxygen (hypoxic), with a low pH and a reducing environment in comparison to non-cancerous cells.[248-250] Hypoxic cancer cells are frequently resistant to conventional cancer therapeutics and thus the development of cobalt(III) prodrugs which can undergo bioreductive activation selectively in hypoxic regions are attractive.[248, 251, 252] Cobalt(III) complexes are generally kinetically inert due to high stabilisation of the d^6 low spin state, whereas cobalt(II) complexes are high spin d^7 and thus more labile and able to undergo ligand exchange. The cobalt(III) complex acts as the inactive prodrug which is reduced to the cobalt(II) species which may itself be reversibly reoxidised to enhance selectivity (**Figure 1.22**).[253, 254] Generally a drug molecule is attached to a cobalt(III) scaffold and released upon reduction, however there are cases where the cobalt(II) complex itself acts as the drug. Ubiquitous reductive enzymes such as cytochrome P450 and xanthine oxidase are thought to be responsible for the bioreduction.[255] However it has been speculated that the rate of reoxidation is more important than the rate of reduction.[256] The cytosolic reduction potential is approximately -200 to -400 mV. An ideal hypoxia activated prodrug should have a biocompatible reduction potential but also a high activation threshold as normal cells may also be hypoxic under certain conditions.[248, 257, 258] The reduction potential, charge, solubility, hydrophobicity and other important biological factors of a cobalt(III) prodrug may be tuned by variations of the ancillary ligands.
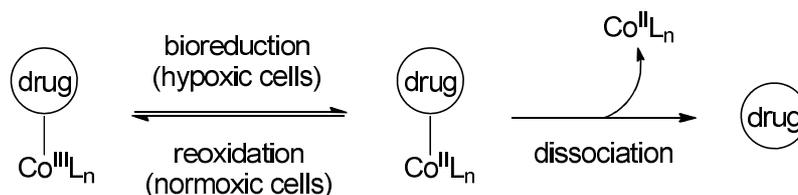
Figure 1.22: Proposed mechanism of action of cobalt(III) prodrugs

The most investigated cobalt complexes for hypoxic activation have included nitrogen mustard ligands; however some studies have been performed with DNA intercalators.[259] Nitrogen mustards are approved anti-cancer drugs which alkylate DNA, form interstrand crosslinks and thereby initiate apoptosis.[260, 261] Cobalt(III) complexes with nitro and amine ligands were found to be effective radiosensitisers for hypoxic cells, with complexes containing acac and nitrogen mustard ligands

particularly effective.[262] The mustard *bis*(2-chloroethyl)amine (bca) was studied (**Figure 1.23a**) because of its deactivation upon coordination to the cobalt(III) centre as the nitrogen lone pair is involved in bonding and may not form the aziridinium ion which is essential for DNA alkylation, upon bioreduction to cobalt(II) the cytotoxic bca is released. The cobalt-bca complex was more active against normoxic cells (normal oxygen levels) compared to hypoxic cells, however this complex was found to circumvent mustard related resistance possibly due to improved cellular uptake.[263]
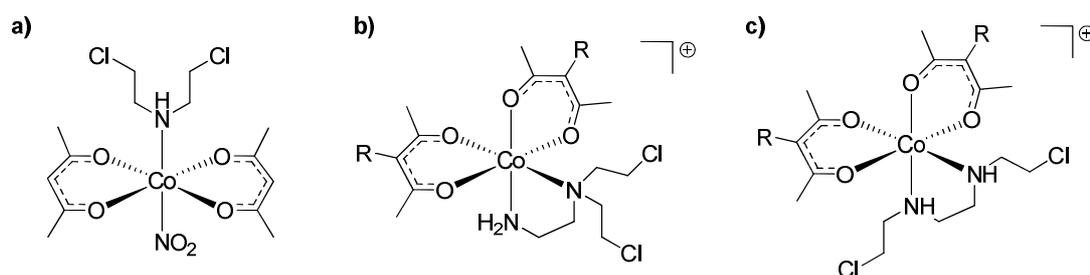
Figure 1.23: Structures of cobalt acac mustard complexes with a) *bis*(2-chloroethyl)amine (bca) b) *N,N-bis*(2-chloroethyl)ethylenediamine (dce) c) *N,N'-bis*(2-chloroethyl)ethylenediamine (bce)

Further cobalt(III) mustard complexes with aziridine groups were investigated; however these were readily reduced with facile release of the cytotoxic ligand.[264, 265] In order to prepare more stable complexes, the bidentate mustards *N,N-bis*(2-chloroethyl)ethylenediamine (dce) and *N,N'-bis*(2-chloroethyl)ethylenediamine (bce) were utilised (**Figure 1.23b** and **Figure 1.23c**). These complexes were able to selectively kill hypoxic cells with cytotoxicity attributed to the free cytotoxic mustard ligand. The dce complexes were more cytotoxic and showed greater hypoxic selectivity than the bce complexes due to the more positive reduction potentials of these complexes and thus more rapid release of the mustard ligand.[265-268] The most effective cobalt-dce complex showed up to 20-fold hypoxic selectivity and was active against tumour spheroids, which have hypoxic centres and thus toxicity is thought to be due to release of dce in the hypoxic core followed by diffusion into the normoxic regions.[264, 267] Mechanistic investigations showed that the mustard ligand was rapidly released independent of oxygen status with no reoxidation of the cobalt(II) species. Therefore the hypoxic selectivity is hypothesised to be due to competition between the cobalt(III) species and oxygen for intracellular reductants.[269] It was suggested that stability of the cobalt(III) complex is an important factor in hypoxic selectivity, however cobalt complexes with more stable tridentate mustard ligands did not display enhanced hypoxic selectivity.[264]

Hypoxic selectivity can be improved by tuning the ancillary acac ligand, with methyl and ethyl acac analogues showing improved selectivity compared to the unsubstituted acac.[267] The effect of replacement of the ancillary acac ligand in the cobalt-dce and bce complexes on hypoxic selectivity was investigated. *Bis*-troplonate and dithiocarbamate complexes were unsuitable due to high reduction potentials leading to complexes which were easily reduced with the facile release of the cytotoxic ligand and thus no hypoxic selectivity.[270, 271] Anionic carbonate and oxalate complexes were less cytotoxic than free dce with lower hypoxic selectivity than the acac complex.[272] Complexes with triazoles have been shown to have structural and redox properties suitable for bioreduction in hypoxia, but these have yet to undergo biological investigation.[273]

Activation of cobalt(III) complexes by reduction in hypoxia can result in a cobalt(II) complex which is active, rather than leading to the release of an active drug. Cobalt(III) complexes with two 2,4-diiodo-6-((pyridine-2-ylmethylamino)methyl) phenol ligands could be reduced in the presence of ascorbic acid to cobalt(II) complexes with the subsequent release of one of the ligands. These cobalt(II) complexes are hypothesised to undergo ligand exchange with amino acid residues at the active site of the 20S and 26S proteasomes leading to inhibition of the proteasomes and induction of apoptosis.[274, 275]

1.7.4 Cobalt chaperones with bioactive ligands

The ability of cobalt(III) to undergo bioreduction has allowed the development of cobalt(III) scaffolds to selectively deliver target drugs to hypoxic sites. Selective delivery of MMP inhibitors to hypoxic cells has been investigated extensively (**Figure 1.24**). MMPs degrade the extracellular matrix and in cancer these enzymes are often overexpressed, facilitating tumour cell invasion and metastasis.[276] Current MMP inhibitors, such as marimastat and batimastat, have been developed as anti-metastatic agents but contain an exposed hydroxamic group which is inherently reactive and prone to undergo side-reactions *in vivo* resulting in poor bioavailability and drug efficacy.[277] Marimastat may coordinate to cobalt through the hydroxamic group which effectively shields the drug from side reactions in normoxic regions, the reduction of cobalt(III) to cobalt(II) in hypoxic regions then allows the selective release of the drug. Tripodal tetradentate ligands, mainly *tris*(2-methylpyridyl)amine (tpa), have been employed as the scaffold to maximise stability.[278, 279] The cobalt(III)-tpa marimastat complex (**Figure 1.24**) suppressed tumour growth in a mouse model to a greater extent than free marimastat, however the complex was

also discovered to potentiate metastasis.[278] In similar complexes with simple alkyl and aryl hydroxamic acids (to act as a model of marimastat) it was found that both the hydroxamate or hydroximate forms could be adopted. These forms are associated with different reduction potentials and thus the ability of these to be released from the complex in a hypoxic environment is also divergent. Different scaffold ligands have been developed to tune hypoxic dependent drug release and also pH dependent release.[279-281]
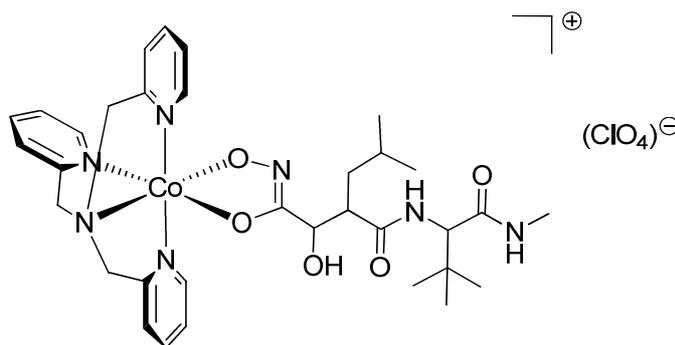
Figure 1.24: Structure of cobalt tpa marimastat complex

Other cobalt scaffolds which have been investigated include cobalt cyclens and cobalt cyclams with a variety of drugs such as 8-hydroxyquinoline and azachloromethylbenzindoline (azaCBI). These complexes are able to release the cytotoxic ligand in hypoxic cells upon exposure to ionising radiation leading to improved selectivity for hypoxic cells over normoxic cells.[282-285] Modification of azaCBI allowed tuning of drug release and thus improved hypoxic selectivity.[286] Cobalt cyclam complexes with azaCBI displayed good cytotoxicity and hypoxic selectivity, however cytotoxicity was not improved with increased concentrations of cytochrome P450 oxidoreductase, a known initiator of other hypoxia-activated drugs.[287]

Cobalt curcumin complexes have recently undergone investigation with a variety of scaffold ligands. Curcumin is a natural product with anti-cancer effects but shows poor bioavailability.[288] Complexes with tpa, phenanthroline and phenolate ligands have all shown improved cellular uptake compared to the free ligand. Photorelease of curcumin following irradiation with visible light is noted leading to improved cytotoxicity in light compared to dark conditions allowing improved cell targeting. The mechanism of these complexes is thought to be through the generation of ROS leading to apoptosis.[289-292]

1.7.5 Cobalamin-drug conjugates

Cobalamin (vitamin B_{12}) is an essential water soluble vitamin which consists of a planar corrin ring coordinating a central cobalt(III) ion with an associated nucleotide derivative (**Figure 1.25**). Cancerous cells, along with other rapidly dividing cells, have a high requirement for cobalamin and thus the formation of cobalamin-drug conjugates has been investigated as a method of selectively targeting cancer cells and thus improve the pharmacokinetic properties of any associated drug conjugate. Conjugation of a drug to cobalamin occurs at four main sites: the β-axial position of the cobalt metal centre, the peripheral propionamide sites on the corrin ring, the 5'-hydroxyl group of the ribose and the phosphate group of the ribose unit (**Figure 1.25**).[293] Following cellular internalisation of the cobalamin-drug conjugate, the bond between cobalamin and the drug is easily broken to allow release of the cytotoxic drug.
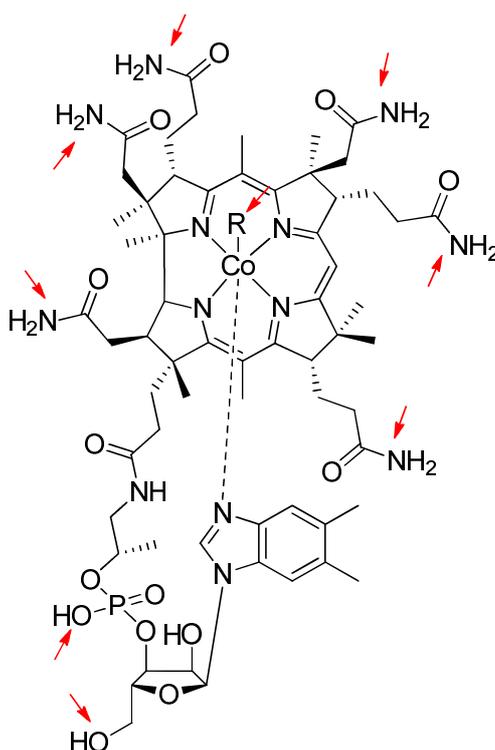
Figure 1.25: Structure of cobalamin including potential conjugation sites (red arrows)

Cobalamin conjugates with known drugs such as chlorambucil and colchicine have shown good anti-cancer activity. The cobalamin-chlorambucil conjugate was as toxic as the free drug; however the colchicine conjugate was found to be less toxic than the free drug. This is possibly due to incomplete release of the drug from the

conjugate due to the nature of the hydrazine linker, designed to enable pH dependent release of the drug.[294, 295] Cobalamin conjugates with cisplatin and cisplatin isomers were active or moderately active against cancer cell lines, however the activity was poorer than free cisplatin possibly due to low cellular accumulation.[296, 297] Conjugates of cobalamin with complexes of toxic metal ions such as gadolinium(III) were cytotoxic and reduced tumour viability, whereas complexes with technetium allowed visualisation through computed tomography (CT) and single-photon emission computed tomography (SPECT).[298, 299] Conjugates of cobalamin with a rhenium carbonyl complex were cytotoxic and allowed visualisation of the cell through fluorescence.[300] The use of cobalamin as a scaffold for controlled photoreactive drug release with red, far-red and near infrared (NIR) light is also under investigation.[301] Small molecules may also be transported as conjugates with cobalamin, for example nitric oxide (NO) in the form nitrosylcobalamin. Nitrosylcobalamin is internalised through endocytosis with the consequential release of NO which may inhibit normal metabolism, inducing DNA damage and apoptosis.[302, 303] *In vivo* studies of nitrosylcobalamin in dogs showed promising improvements in tumour size following treatment with low systemic toxicity.[304]

1.8 Metal picolinamide complexes

Complexes incorporating picolinamide ligands have been reported with a wide variety of metals including copper, nickel, iron, manganese, zinc, cadmium, cobalt, ruthenium, osmium and the lanthanides amongst others.[170, 305-311] The McGowan group has extensively researched picolinamide complexes with ruthenium, rhodium, iridium, titanium and copper for application in catalysis and drug discovery.[170, 171, 312-314]

Ruthenium, rhodium and iridium arene complexes with picolinamide ligands have been investigated for their anti-cancer potential within the McGowan group. Picolinamide ligands within ruthenium *p*-cymene complexes were found to adopt different coordination modes (*N,N* or *N,O* bound) depending upon the nature of the ligand substituent modulated by temperature and pH (**Figure 1.26a** and **Figure 1.26b**). Neutral complexes with *N,N* bound picolinamide underwent rapid hydrolysis, rapid binding to guanine and significant cytotoxicity ($IC_{50} < 25$ μM); in contrast monocationic complexes with *N,O* bound picolinamide underwent slow hydrolysis, did not bind to DNA and were non-cytotoxic.[310] The positioning, number and nature

of the substituents on the picolinamide ligand was found to have a significant effect on cytotoxicity.[170] Analogous monocationic ruthenium *p*-cymene complexes with N-methyl picolinamide ligands (*N,O* bound) are non-toxic.[315] Rhodium and iridium Cp* complexes with picolinamide ligands (**Figure 1.26c**) showed moderate anti-cancer activity (IC$_{50}$ = 30 - 80 μM), with improvements in activity when the picolinamide ligand is substituted for a ketoiminate ligand (IC$_{50}$ = 3 - 5 μM).[171] Rhodium and iridium picolinamide complexes with functionalised Cp* arene containing hydroxyl tethered arms of various carbon chain lengths displayed cytotoxicity (IC$_{50}$ = 50 - 130 μM) against a number of different cell lines (**Figure 1.26d**). Cytotoxicity improved proportionally with carbon chain length, with the most promising results for the 14-carbon tethered Cp* rhodium and iridium picolinamide complexes. The mechanism of action of these complexes may involve disruption of cell anti-oxidant function through inhibition of thioredoxin reductase.[316]
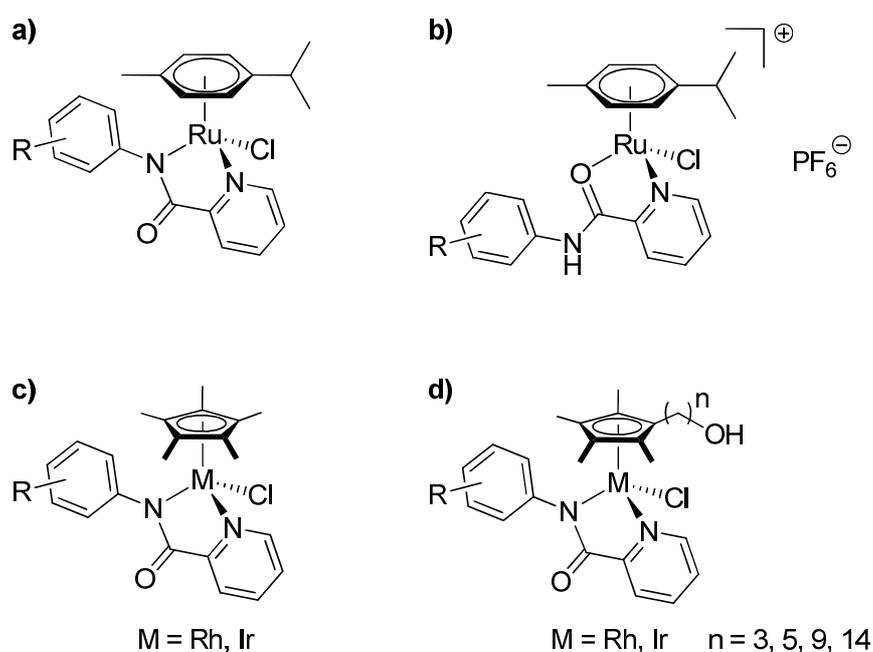
Figure 1.26: Metal arene picolinamide complexes a) neutral ruthenium *p*-cymene complexes with *N,N*-bound picolinamide b) monocationic ruthenium *p*-cymene complexes with *N,O*-bound picolinamide c) rhodium and iridium Cp* picolinamide complexes d) rhodium and iridium hydroxyl tethered Cp* picolinamide complexes

Ruthenium and rhodium coordination complexes with picolinamide ligands have also been investigated for their anti-cancer potential within the McGowan group. These complexes consist of two picolinamide ligands, one *N,N* bound and one *N,O* bound, and two labile halide ligands attached to a ruthenium(III) or rhodium(III)

central ion (**Figure 1.27**). Ruthenium and rhodium complexes with labile chloride ligands formed a mixture of three geometric isomers: *cis-trans-cis*, *cis-cis-cis* and *trans-trans-trans*. However, when iodide ligands are present only the *trans-trans-trans* isomer is formed. This is thought to be due to the larger ionic radius of the iodide, compared to the chloride ion, which means that the iodide ions preferentially adopt the *trans* position to minimise electronic overlap. Ruthenium and rhodium complexes with iodide ligands displayed improved cytotoxicity (IC$_{50}$ = 1 - 40 µM) compared to the complexes with chloride ligands (IC$_{50}$ = 5 - 90 µM). In addition, substitutions in the *meta* and *para* positions of the picolinamide aryl ring were associated with improved cytotoxicity. In the chloride complexes, improved cytotoxicity was associated with increased hydrolysis rates. Whereas in the iodide complexes, improved cytotoxicity was associated with decreased hydrolysis rates.[312]

cis-trans-cis   *cis-cis-cis*   *trans-trans-trans*
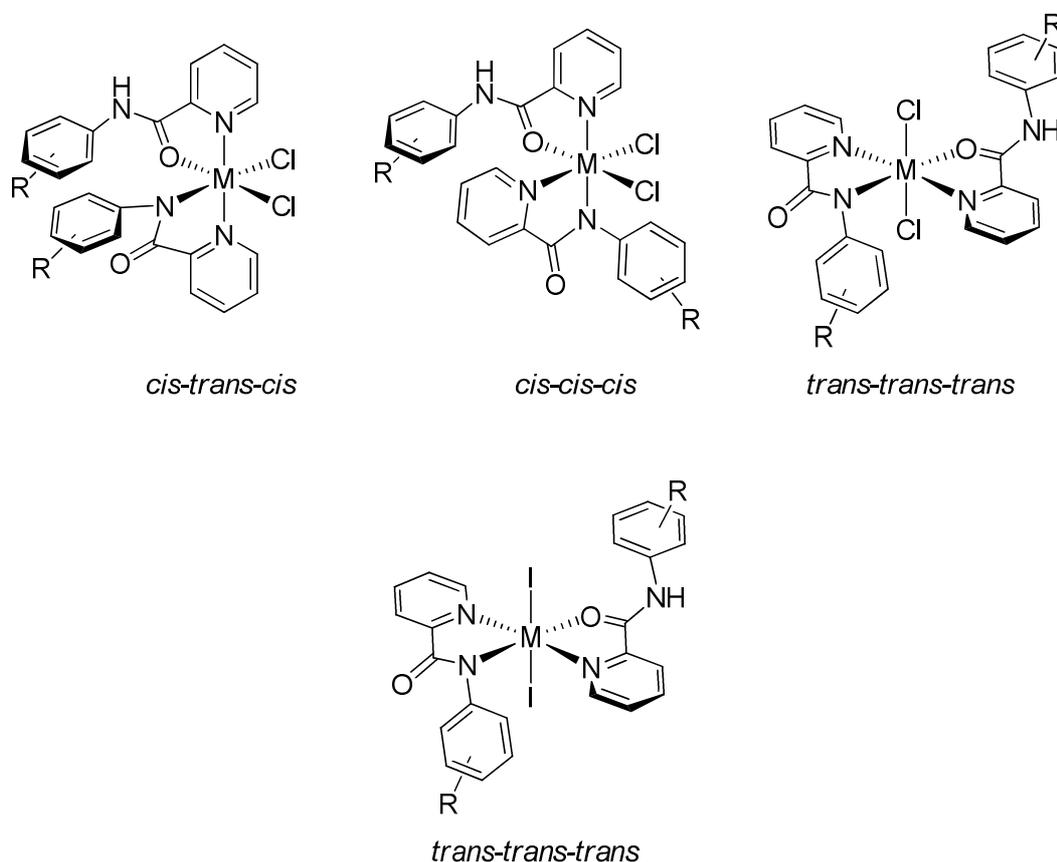
trans-trans-trans

Figure 1.27: Metal *bis*-picolinamide *bis*-halide complexes. Geometric (*cis* / *trans*) descriptors are designated in the order: halide ligand, picolinamide pyridyl ring, picolinamide amide group. M = Ru, Rh

1.9 Objectives

The objective of this research was the exploration of the chemistry and biology of a variety of cobalt picolinamide complexes with different structural features. Metal picolinamide complexes previously investigated contain a number of elements which are thought to be important for anti-cancer activity (**Figure 1.28**). The planar aromatic pyridyl and aryl rings of the picolinamide ligand provide a potential site for π-π stacking interactions with nucleobases within DNA. The acidic amide proton of the picolinamide ligand provides a potential hydrogen bonding site for interactions with biomolecules. The substituents on the picolinamide aryl ring can affect the hydrophobicity and cellular uptake of the complex and can affect the anti-cancer activity. The presence of a labile halide or pseudohalide ligand allows the possibility of activation by ligand exchange to form more reactive species which may then interact with target biomolecules.
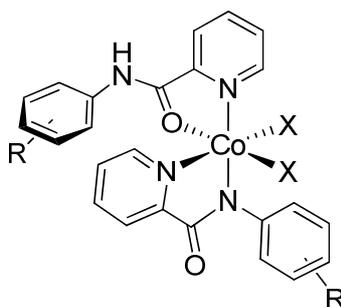
Figure 1.28: Potential cobalt picolinamide complexes including sites of potential variation

There have only been a few reported cobalt picolinamide complexes with even fewer having undergone biological investigation. There are a number of features of cobalt picolinamide complexes which may be varied in order to fully explore the potential of these complexes. The oxidation state of the central cobalt ion may be varied as cobalt(II) and cobalt(III) complexes are stable in biological systems. The number of picolinamide ligands and the nature of the ligand binding to the metal (*N,N* or *N,O* coordination) can be varied and will affect the overall charge of the complex and the number of possible isomers. The substituents present on the picolinamide aryl ring can be varied to investigate how differing electronic and steric groups affect the biological activity. Additional ligands may also be varied from labile halide ligands to more stable alternative bidentate ligands. This research aimed to investigate the importance of all of these different structural variations within cobalt picolinamide complexes with respect to the biological activity of the complexes.

1.10 References

1.   P. D. Sasieni, J. Shelton, N. Ormiston-Smith, C. S. Thomson and P. B. Silcocks, *British Journal of Cancer*, 2011, **105**, 460-465.

2.   Cancer Research UK, *Cancer mortality for all cancers combined*, http://www.cancerresearchuk.org/health-professional/cancer-statistics/mortality, Accessed 06/02/2017.

3.   A. Aggarwal and R. Sullivan, *Journal of Cancer Policy*, 2014, **2**, 31-39.

4.   Cancer Research UK, *Cancer incidence statistics*, http://www.cancerresearchuk.org/health-professional/cancer-statistics/incidence, Accessed 06/02/2017.

5.   A. Jemal, F. Bray, M. M. Center, J. Ferlay, E. Ward and D. Forman, *Ca- A Cancer Journal for Clinicians*, 2011, **61**, 69-90.

6.   Cancer Research UK, *Worldwide cancer statistics*, http://www.cancerresearchuk.org/health-professional/cancer-statistics/worldwide-cancer, Accessed 06/02/2017.

7.   World Health Organisation, *Cancer*, http://www.who.int/mediacentre/factsheets/fs297/en/, Accessed 06/02/2017.

8.   A. G. Knudson, *Proceedings of the National Academy of Sciences of the United States of America*, 1971, **68**, 820-823.

9.   P. Boffetta and F. Nyberg, *British Medical Bulletin*, 2003, **68**, 71-94.

10.   D. Hanahan and R. A. Weinberg, *Cell*, 2000, **100**, 57-70.

11.   D. Hanahan and R. A. Weinberg, *Cell*, 2011, **144**, 646-674.

12.   S. Mercadante, V. Gebbia, A. Marrazzo and S. Filosto, *Cancer Treatment Reviews*, 2000, **26**, 303-311.

13.   H. Suzuki, A. Asakawa, H. Amitani, N. Nakamura and A. Inui, *Journal of Gastroenterology*, 2013, **48**, 574-594.

14.   M. von Elstermann, *Nature Reviews Cancer*, 2008, **8**, 410-411.

15.   A. Urruticoechea, R. Alemany, J. Balart, A. Villanueva, F. Vinals and G. Capella, *Current Pharmaceutical Design*, 2010, **16**, 3-10.

16.   Institute of Medicine, eds. C. M. Pechura and D. P. Rall, National Acadamy Press, Washington, Editon edn., 1993.

17.   V. T. DeVita, Jr. and E. Chu, *Cancer Research*, 2008, **68**, 8643-8653.

18.   M. Frezza, S. Hindo, D. Chen, A. Davenport, S. Schmitt, D. Tomco and Q. P. Dou, *Current Pharmaceutical Design*, 2010, **16**, 1813-1825.

19.   C. Orvig and M. J. Abrams, *Chemical Reviews*, 1999, **99**, 2201-2203.

20.   F. P. Dwyer, E. C. Gyarfas, W. P. Rogers and J. H. Koch, *Nature*, 1952, **170**, 190-191.

21.   D. Gaynor and D. M. Griffith, *Dalton Transactions*, 2012, **41**, 13239-13257.

22.   C. A. Lipinski, F. Lombardo, B. W. Dominy and P. J. Feeney, *Advanced Drug Delivery Reviews*, 2001, **46**, 3-26.

23.   P. Yang and M. L. Guo, *Coordination Chemistry Reviews*, 1999, **185-6**, 189-211.

24.   N. Farrell, L. R. Kelland, J. D. Roberts and M. Vanbeusichem, *Cancer Research*, 1992, **52**, 5065-5072.

25.   L. R. Kelland, C. F. J. Barnard, K. J. Mellish, M. Jones, P. M. Goddard, M. Valenti, A. Bryant, B. A. Murrer and K. R. Harrap, *Cancer Research*, 1994, **54**, 5618-5622.

26.   A. L. Noffke, A. Habtemariam, A. M. Pizarro and P. J. Sadler, *Chemical Communications*, 2012, **48**, 5219-5246.

27.   B. Rosenberg, L. Vancamp, J. E. Trosko and V. H. Mansour, *Nature*, 1969, **222**, 385-386.

28.   O. R. Allen, R. J. Knox and P. C. McGowan, *Dalton Transactions*, 2008, 1293-1295.

29.   A. Cuin, A. C. Massabni, G. A. Pereira, C. Q. Fujimura Leite, F. R. Pavan, R. Sesti-Costa, T. A. Heinrich and C. M. Costa-Neto, *Biomedicine & Pharmacotherapy*, 2011, **65**, 334-338.

30.   B. Desoize, *Anticancer Research*, 2004, **24**, 1529-1544.

31.   Z. Liu, A. Habtemariam, A. M. Pizarro, S. A. Fletcher, A. Kisova, O. Vrana, L. Salassa, P. C. A. Bruijnincx, G. J. Clarkson, V. Brabec and P. J. Sadler, *Journal of Medicinal Chemistry*, 2011, **54**, 3011-3026.

32.   I. Ott and R. Gust, *Archiv Der Pharmazie*, 2007, **340**, 117-126.

33.   C. Santini, M. Pellei, V. Gandin, M. Porchia, F. Tisato and M. Marzano, *Chemical Reviews*, 2014, **114**, 815-862.

34.   S. H. van Rijt, A. F. A. Peacock, R. D. L. Johnstone, S. Parsons and P. J. Sadler, *Inorganic Chemistry*, 2009, **48**, 1753-1762.

35.   A. R. Kapdi and I. J. S. Fairlamb, *Chemical Society Reviews*, 2014, **43**, 4751-4777.

36.   M. Nath, M. Vats and P. Roy, *European Journal of Medicinal Chemistry*, 2013, **59**, 310-321.

37.   E. Wong and C. M. Giandomenico, *Chemical Reviews*, 1999, **99**, 2451-2466.

38.   B. Rosenberg, L. Vancamp and T. Krigas, *Nature*, 1965, **205**, 698-699.

39.   B. Rosenberg and L. Vancamp, *Cancer Research*, 1970, **30**, 1799-1802.

40.   E. Blanchard, *Journal of Solid Tumors*, 2012, **2**, 26-33.

41.   National Cancer Institute at the National Institutes of Health, *Cancer drug information   -   cisplatin*, http://www.cancer.gov/cancertopics/druginfo/cisplatin, Accessed 15/02/2017.

42.   J. J. Liu, J. Lu and M. J. McKeage, *Current Cancer Drug Targets*, 2012, **12**, 962-986.

43.   B. Crossley, PhD Thesis, The University of Leeds, 2011.

44.   A. Eastman, *Pharmacology & Therapeutics*, 1987, **34**, 155-166.

45.   A. E. Egger, C. G. Hartinger, H. Ben Hamidane, Y. O. Tsybin, B. K. Keppler and P. J. Dyson, *Inorganic Chemistry*, 2008, **47**, 10626-10633.

46.   A. M. J. Fichtinger-Schepman, J. L. Vanderveer, J. H. J. Denhartog, P. H. M. Lohman and J. Reedijk, *Biochemistry*, 1985, **24**, 707-713.

47.   E. R. Jamieson and S. J. Lippard, *Chemical Reviews*, 1999, **99**, 2467-2498.

48.   J. J. Roberts and F. Friedlos, *Pharmacology & Therapeutics*, 1987, **34**, 215-246.

49.   C. A. Rabik and M. E. Dolan, *Cancer Treatment Reviews*, 2007, **33**, 9-23.

50.   M. A. Fuertes, J. Castilla, C. Alonso and J. M. Perez, *Current Medicinal Chemistry - Anti-Cancer Agents*, 2002, **2**, 539-551.

51.   T. W. Hambley, *Journal of the Chemical Society-Dalton Transactions*, **2001**, 2711-2718.

52.   A. Basu and S. Krishnamurthy, *Journal of Nucleic Acids*, 2010, **2010**, Article ID: 201367.

53.   R. A. Alderden, M. D. Hall and T. W. Hambley, *Journal of Chemical Education*, 2006, **83**, 728-734.

54.   A. Gelasco and S. J. Lippard, *Biochemistry*, 1998, **37**, 9230-9239.

55.   N. J. Wheate, S. Walker, G. E. Craig and R. Oun, *Dalton Transactions*, 2010, **39**, 8113-8127.

56.   A. I. Ivanov, J. Christodoulou, J. A. Parkinson, K. J. Barnham, A. Tucker, J. Woodrow and P. J. Sadler, *Journal of Biological Chemistry*, 1998, **273**, 14721-14730.

57.   R. C. Dolman, G. B. Deacon and T. W. Hambley, *Journal of Inorganic Biochemistry*, 2002, **88**, 260-267.

58.   R. F. Borch and M. E. Pleasants, *Proceedings of the National Academy of Sciences of the United States of America*, 1979, **76**, 6611-6614.

59.   J. P. Fillastre and G. Raguenezviotte, *Toxicology Letters*, 1989, **46**, 163-175.

60.   N. Pabla and Z. Dong, *Oncotarget*, 2012, **3**, 107-111.

61.   A. S. Jaggi and N. Singh, *Toxicology*, 2012, **291**, 1-9.

62.   P. R. Brock, K. R. Knight, D. R. Freyer, K. C. M. Campbell, P. S. Steyger, B. W. Blakley, S. R. Rassekh, K. W. Chang, B. J. Fligor, K. Rajput, M. Sullivan and E. A. Neuwelt, *Journal of Clinical Oncology*, 2012, **30**, 2408-2417.

63.   H. H. W. Chen and M. T. Kuo, *Metal-Based Drugs*, 2010, Article No.: 430939.

64.   Z. H. Siddik, *Oncogene*, 2003, **22**, 7265-7279.

65.   D. Wang and S. J. Lippard, *Nature Reviews Drug Discovery*, 2005, **4**, 307-320.

66.   U. Frey, J. D. Ranford and P. J. Sadler, *Inorganic Chemistry*, 1993, **32**, 1333-1340.

67.   J. Reedijk, *Chemical Communications*, 1996, **32**, 801-806.

68.   T. Yasumasu, T. Ueda, J. Uozumi, Y. Mihara and J. Kumazawa, *Pharmacology & Toxicology*, 1992, **70**, 143-147.

69.   L. R. Kelland, *Critical Reviews in Oncology/Hematology*, 1993, **15**, 191-219.

70.   National Cancer Institute at the National Institutes of Health, *Cancer drug information   -   carboplatin*, http://www.cancer.gov/cancertopics/druginfo/carboplatin, Accessed 15/02/2017.

71.   National Cancer Institute at the National Institutes of Health, *Cancer drug information   -   oxaliplatin*, http://www.cancer.gov/cancertopics/druginfo/oxaliplatin, Accessed 15/02/2017.

72.   S. Giacchetti, B. Perpoint, R. Zidani, N. Le Bail, R. Faggiuolo, C. Focan, P. Chollet, J. F. Llory, Y. Letourneau, B. Coudert, F. Bertheaut-Cvitkovic, D. Larregain-Fournier, A. Le Rol, S. Walter, R. Adam, J. L. Misset and F. Levi, *Journal of Clinical Oncology*, 2000, **18**, 136-147.

73.   E. Raymond, S. Faivre, S. Chaney, J. Woynarowski and E. Cvitkovic, *Molecular Cancer Therapeutics*, 2002, **1**, 227-235.

74.   T. C. Johnstone, *Polyhedron*, 2014, **67**, 429-435.

75.   B. Spingler, D. A. Whittington and S. J. Lippard, *Inorganic Chemistry*, 2001, **40**, 5596-5602.

76.   A. Bergamo and G. Sava, *Chemical Society Reviews*, 2015, **44**, 8818-8835.

77.   T. C. Johnstone, K. Suntharalingam and S. J. Lippard, *Chemical Reviews*, 2016, **116**, 3436-3486.

78.   E. Musgrove, C. Rugg, I. Taylor and D. Hedley, *Journal of Cellular Physiology*, 1984, **118**, 6-12.

79.   K. C. Reed and F. L. Bygrave, *Biochemical Journal*, 1974, **140**, 143-155.

80.   M. J. Clarke, F. C. Zhu and D. R. Frasca, *Chemical Reviews*, 1999, **99**, 2511-2533.

81.   G. Sava, I. Capozzi, K. Clerici, G. Gagliardi, E. Alessio and G. Mestroni, *Clinical & Experimental Metastasis*, 1998, **16**, 371-379.

82.   M. Bacac, A. C. G. Hotze, K. van der Schilden, J. G. Haasnoot, S. Pacor, E. Alessio, G. Sava and J. Reedijk, *Journal of Inorganic Biochemistry*, 2004, **98**, 402-412.

83.   G. Mestroni, E. Alessio, G. Sava, S. Pacor, M. Coluccia and A. Boccarelli, *Metal-Based Drugs*, 1994, **1**, 41-63.

84.   G. Sava, A. Bergamo, S. Zorzet, B. Gava, C. Casarsa, M. Cocchietto, A. Furlani, V. Scarcia, B. Serli, E. Iengo, E. Alessio and G. Mestroni, *European Journal of Cancer*, 2002, **38**, 427-435.

85.   A. Bergamo, R. Gagliardi, V. Scarcia, A. Furlani, E. Alessio, G. Mestroni and G. Sava, *Journal of Pharmacology and Experimental Therapeutics*, 1999, **289**, 559-564.

86.   G. Sava, S. Pacor, E. Alessio, G. Mestroni, R. Gagliardi, M. Cocchietto and M. Coluccia, *Drugs of the Future*, 1993, **18**, 894-900.

87.   G. Sava, S. Zorzet, C. Turrin, F. Vita, M. Soranzo, G. Zabucchi, M. Cocchietto, A. Bergamo, S. DiGiovine, G. Pezzoni, L. Sartor and S. Garbisa, *Clinical Cancer Research*, 2003, **9**, 1898-1905.

88.   S. Leijen, S. A. Burgers, P. Baas, D. Pluim, M. Tibben, E. van Werkhoven, E. Alessio, G. Sava, J. H. Beijnen and J. H. M. Schellens, *Investigational new drugs*, 2015, **33**, 201-214.

89.   P. F. Salas, C. Herrmann and C. Orvig, *Chemical Reviews*, 2013, **113**, 3450-3492.

90.   F. Frausin, M. Cocchietto, A. Bergamo, V. Searcia, A. Furlani and G. Sava, *Cancer Chemotherapy and Pharmacology*, 2002, **50**, 405-411.

91.   A. A. Hostetter, M. L. Miranda, V. J. DeRose and K. L. M. Holman, *Journal of Biological Inorganic Chemistry*, 2011, **16**, 1177-1185.

92.   D. Pluim, R. van Waardenburg, J. H. Beijnen and J. H. M. Schellens, *Cancer Chemotherapy and Pharmacology*, 2004, **54**, 71-78.

93.   L. Morbidelli, S. Donnini, S. Filippi, L. Messori, F. Piccioli, P. Orioli, G. Sava and M. Ziche, *British Journal of Cancer*, 2003, **88**, 1484-1491.

94.   G. Sava, I. Capozzi, A. Bergamo, R. Gagliardi, M. Cocchietto, L. Masiero, M. Onisto, E. Alessio, G. Mestroni and S. Garbisa, *International Journal of Cancer*, 1996, **68**, 60-66.

95.   B. Gava, S. Zorzet, P. Spessotto, M. Cocchietto and G. Sava, *Journal of Pharmacology and Experimental Therapeutics*, 2006, **317**, 284-291.

96.   G. Sava, F. Frausin, M. Cocchietto, F. Vita, E. Podda, P. Spessotto, A. Furlani, V. Scarcia and G. Zabucchi, *European Journal of Cancer*, 2004, **40**, 1383-1396.

97.   A. Bergamo, C. Gaiddon, J. H. M. Schellens, J. H. Beijnen and G. Sava, *Journal of Inorganic Biochemistry*, 2012, **106**, 90-99.

98.   F. T. Garzon, M. R. Berger, B. K. Keppler and D. Schmahl, *Cancer Chemotherapy and Pharmacology*, 1987, **19**, 347-349.

99.   B. K. Keppler, M. Henn, U. M. Juhl, M. R. Berger, R. Niebl and F. E. Wagner, in *Ruthenium and other non-platinum metal complexes in cancer chemotherapy*, Springer-Verlag Berlin Heidelberg, Editon edn., 1989, vol. First.

100.   W. Peti, T. Pieper, M. Sommer, B. K. Keppler and G. Giester, *European Journal of Inorganic Chemistry*, 1999, 1551-1555.

101.   R. Trondl, P. Heffeter, C. R. Kowol, M. A. Jakupec, W. Berger and B. K. Keppler, *Chemical Science*, 2014, **5**, 2925-2932.

102.   C. G. Hartinger, M. A. Jakupec, S. Zorbas-Seifried, M. Groessl, A. Egger, W. Berger, H. Zorbas, P. J. Dyson and B. K. Keppler, *Chemistry & Biodiversity*, 2008, **5**, 2140-2155.

103.   P. Heffeter, K. Bock, B. Atil, M. A. R. Hoda, W. Korner, C. Bartel, U. Jungwirth, B. K. Keppler, M. Micksche, W. Berger and G. Koellensperger, *Journal of Biological Inorganic Chemistry*, 2010, **15**, 737-748.

104.   J. M. Rademaker-Lakhai, D. van den Bongard, D. Pluim, J. H. Beijnen and J. H. M. Schellens, *Clinical Cancer Research*, 2004, **10**, 3717-3727.

105.   F. Piccioli, S. Sabatini, L. Messori, P. Orioli, C. G. Hartinger and B. K. Keppler, *Journal of Inorganic Biochemistry*, 2004, **98**, 1135-1142.

106.   A. R. Timerbaev, L. S. Foteeva, A. V. Rudnev, J. K. Abramski, K. Polec-Pawlak, C. G. Hartinger, M. Jarosz and B. K. Keppler, *Electrophoresis*, 2007, **28**, 2235-2240.

107.   M. J. Clarke, *Coordination Chemistry Reviews*, 2003, **236**, 207.

108.   A. Egger, V. B. Arion, E. Reisner, B. Cebrian-Losantos, S. Shova, G. Trettenhahn and B. K. Keppler, *Inorganic Chemistry*, 2005, **44**, 122-132.

109.   M. Groessl, Y. O. Tsybin, C. G. Hartinger, B. K. Keppler and P. J. Dyson, *Journal of Biological Inorganic Chemistry*, 2010, **15**, 677-688.

110.   P. Heffeter, U. Jungwirth, M. Jakupec, C. Hartinger, M. Galanski, L. Elbling, M. Micksche, B. Keppler and W. Berger, *Drug Resistance Updates*, 2008, **11**, 1-16.

111.   S. Kapitza, M. A. Jakupec, M. Uhl, B. K. Keppler and B. Marian, *Cancer Letters*, 2005, **226**, 115-121.

112.   B. S. Murray, M. V. Babak, C. G. Hartinger and P. J. Dyson, *Coordination Chemistry Reviews*, 2016, **306**, 86-114.

113.   C. Scolaro, A. B. Chaplin, C. G. Hartinger, A. Bergamo, M. Cocchietto, B. K. Keppler, G. Sava and P. J. Dyson, *Dalton Transactions*, 2007, 5065-5072.

114.   C. S. Allardyce, P. J. Dyson, D. J. Ellis and S. L. Heath, *Chemical Communications*, 2001, 1396-1397.

115.   C. Scolaro, A. Bergamo, L. Brescacin, R. Delfino, M. Cocchietto, G. Laurenczy, T. J. Geldbach, G. Sava and P. J. Dyson, *Journal of Medicinal Chemistry*, 2005, **48**, 4161-4171.

116.   A. Bergamo, A. Masi, P. J. Dyson and G. Sava, *International Journal of Oncology*, 2008, **33**, 1281-1289.

117.   B. Wu, M. S. Ong, M. Groessl, Z. Adhireksan, C. G. Hartinger, P. J. Dyson and C. A. Davey, *Chem.-Eur. J.*, 2011, **17**, 3562-3566.

118.   C. G. Hartinger, A. Casini, C. Duhot, Y. O. Tsybin, L. Messori and P. J. Dyson, *Journal of Inorganic Biochemistry*, 2008, **102**, 2136-2141.

119.   A. Casini, G. Mastrobuoni, W. H. Ang, C. Gabbiani, G. Pieraccini, G. Moneti, P. J. Dyson and L. Messori, *Chemmedchem*, 2007, **2**, 631-635.

120.   A. Casini, C. Gabbiani, E. Michelucci, G. Pieraccini, G. Moneti, P. J. Dyson and L. Messori, *Journal of Biological Inorganic Chemistry*, 2009, **14**, 761-770.

121.   A. Casini, A. Karotki, C. Gabbiani, F. Rugi, M. Vasak, L. Messori and P. J. Dyson, *Metallomics*, 2009, **1**, 434-441.

122.   A. Casini, C. Gabbiani, F. Sorrentino, M. P. Rigobello, A. Bindoli, T. J. Geldbach, A. Marrone, N. Re, C. G. Hartinger, P. J. Dyson and L. Messori, *Journal of Medicinal Chemistry*, 2008, **51**, 6773-6781.

123.   F. Mendes, M. Groessl, A. A. Nazarov, Y. O. Tsybin, G. Sava, I. Santos, P. J. Dyson and A. Casini, *Journal of Medicinal Chemistry*, 2011, **54**, 2196-2206.

124.   W. S. Sheldrick and S. Heeb, *Inorganica Chimica Acta*, 1990, **168**, 93-100.

125.   R. E. Morris, R. E. Aird, P. D. Murdoch, H. M. Chen, J. Cummings, N. D. Hughes, S. Parsons, A. Parkin, G. Boyd, D. I. Jodrell and P. J. Sadler, *Journal of Medicinal Chemistry*, 2001, **44**, 3616-3621.

126.   Y. K. Yan, M. Melchart, A. Habtemariam and P. J. Sadler, *Chemical Communications*, 2005, **41**, 4764-4776.

127.   S. J. Dougan and P. J. Sadler, *Chimia*, 2007, **61**, 704-715.

128.   L. L. Parker, S. M. Lacy, L. J. Farrugia, C. Evans, D. J. Robins, C. C. O'Hare, J. A. Hartley, M. Jaffar and I. J. Stratford, *Journal of Medicinal Chemistry*, 2004, **47**, 5683-5689.

129.   A. Habtemariam, M. Melchart, R. Fernandez, S. Parsons, I. D. H. Oswald, A. Parkin, F. P. A. Fabbiani, J. E. Davidson, A. Dawson, R. E. Aird, D. I. Jodrell and P. J. Sadler, *Journal of Medicinal Chemistry*, 2006, **49**, 6858-6868.

130.   R. Fernandez, M. Melchart, A. Habtemariam, S. Parsons and P. J. Sadler, *Chem.-Eur. J.*, 2004, **10**, 5173-5179.

131.   F. Wang, H. M. Chen, S. Parsons, L. D. H. Oswald, J. E. Davidson and P. J. Sadler, *Chem.-Eur. J.*, 2003, **9**, 5810-5820.

132.   H. M. Chen, J. A. Parkinson, R. E. Morris and P. J. Sadler, *Journal of the American Chemical Society*, 2003, **125**, 173-186.

133.   M. H. Baik, R. A. Friesner and S. J. Lippard, *Journal of the American Chemical Society*, 2003, **125**, 14082-14092.

134.   R. E. Aird, J. Cummings, A. A. Ritchie, M. Muir, R. E. Morris, H. Chen, P. J. Sadler and D. I. Jodrell, *British Journal of Cancer*, 2002, **86**, 1652-1657.

135.   J. L. Bear, H. B. Gray, L. Rainen, I. M. Chang, R. Howard, G. Serio and A. P. Kimball, *Cancer Chemotherapy Reports Part 1*, 1975, **59**, 611-620.

136.   A. Erck, L. Rainen, Whileyma.J, I. M. Chang, A. P. Kimball and J. Bear, *Proceedings of the Society for Experimental Biology and Medicine*, 1974, **145**, 1278-1283.

137.   L. M. Hall, R. J. Speer and H. J. Ridgway, *Journal of Clinical Hematology and Oncology*, 1980, **10**, 25-28.

138.   R. A. Howard, E. Sherwood, A. Erck, A. P. Kimball and J. L. Bear, *Journal of Medicinal Chemistry*, 1977, **20**, 943-946.

139.   P. N. Rao, M. L. Smith, S. Pathak, R. A. Howard and J. L. Bear, *Journal of the National Cancer Institute*, 1980, **64**, 905-912.

140.   R. A. Howard, T. G. Spring and J. L. Bear, *Cancer Research*, 1976, **36**, 4402-4405.

141.   E. Tselepikalouli and N. Katsaros, *Journal of Inorganic Biochemistry*, 1990, **40**, 95-102.

142.   B. P. Esposito, S. B. Zyngier, A. R. de Souza and R. Najjar, *Metal-Based Drugs*, 1997, **4**, 333-338.

143.   B. P. Esposito, S. B. Zyngier, R. Najjar, R. P. Paes, S. M. Ykko Ueda and J. C. A. Barros, *Metal-Based Drugs*, 1999, **6**, 17-18.

144.   R. Chibber, I. J. Stratford, P. Oneill, P. W. Sheldon, I. Ahmed and B. Lee, *International Journal of Radiation Biology*, 1985, **48**, 513-524.

145.   D. G. Craciunescu, C. Molina, E. Parrondo-Iglesias, M. P. Alonso, C. Lorenzo Molina, J. C. Doadrio-Villarejo, M. T. Gutierrez-Rios and M. I. De Frutos, *Anales de la Real Academia de Farmacia*, 1991, **57**, 15-36.

146.   G. Mestroni, E. Alessio, A. S. Santi, S. Geremia, A. Bergamo, G. Sava, A. Boccarelli, A. Schettino and M. Coluccia, *Inorganica Chimica Acta*, 1998, **273**, 62-71.

147.   S. Pacor, G. Sava, G. Mestroni and E. Alessio, *Pharmacological Research*, 1992, **25**, 73-74.

148.   A. B. Bikhazi, A. Salameh, M. M. Elkasti and R. A. Awar, *Comparative Biochemistry and Physiology C-Pharmacology Toxicology & Endocrinology*, 1995, **111**, 423-427.

149.   L. Salassa, *European Journal of Inorganic Chemistry*, 2011, 4931-4947.

150.   M. Dobroschke, Y. Geldmacher, I. Ott, M. Harlos, L. Kater, L. Wagner, R. Gust, W. S. Sheldrick and A. Prokop, *Chemmedchem*, 2009, **4**, 177-187.

151.   Y. Geldmacher, I. Kitanovic, H. Alborzinia, K. Bergerhoff, R. Rubbiani, P. Wefelmeier, A. Prokop, R. Gust, I. Ott, S. Wolfl and W. S. Sheldrick, *Chemmedchem*, 2011, **6**, 429-439.

152.   Y. Geldmacher, K. Splith, I. Kitanovic, H. Alborzinia, S. Can, R. Rubbiani, M. A. Nazif, P. Wefelmeier, A. Prokop, I. Ott, S. Wolfl, I. Neundorf and W. S. Sheldrick, *Journal of Biological Inorganic Chemistry*, 2012, **17**, 631-646.

153.   M. Harlos, I. Ott, R. Gust, H. Alborzinia, S. Wolfl, A. Kromm and W. S. Sheldrick, *Journal of Medicinal Chemistry*, 2008, **51**, 3924-3933.

154.   C.-H. Leung, H. Yang, V. P.-Y. Ma, D. S.-H. Chan, H.-J. Zhong, Y.-W. Li, W.-F. Fong and D.-L. Ma, *MedChemComm*, 2012, **3**, 696-698.

155.   T. A. Soucy, P. G. Smith, M. A. Milhollen, A. J. Berger, J. M. Gavin, S. Adhikari, J. E. Brownell, K. E. Burke, D. P. Cardin, S. Critchley, C. A. Cullis, A. Doucette, J. J. Garnsey, J. L. Gaulin, R. E. Gershman, A. R. Lublinsky, A. McDonald, H. Mizutani, U. Narayanan, E. J. Olhava, S. Peluso, M. Rezaei, M. D. Sintchak, T. Talreja, M. P. Thomas, T. Traore, S. Vyskocil, G. S. Weatherhead, J. Yu, J. Zhang, L. R. Dick, C. F. Claiborne, M. Rolfe, J. B. Bolen and S. P. Langston, *Nature*, 2009, **458**, U732-U767.

156.   B. Y.-W. Man, H.-M. Chan, C.-H. Leung, D. S.-H. Chan, L.-P. Bai, Z.-H. Jiang, H.-W. Li and D.-L. Ma, *Chemical Science*, 2011, **2**, 917-921.

157.   M. A. Scharwitz, I. Ott, R. Gust, A. Kromm and W. S. Sheldrick, *Journal of Inorganic Biochemistry*, 2008, **102**, 1623-1630.

158.   Y. Geldmacher, R. Rubbiani, P. Wefelmeier, A. Prokop, I. Ott and W. S. Sheldrick, *Journal of Organometallic Chemistry*, 2011, **696**, 1023-1031.

159.   B. A. Jackson, V. Y. Alekseyev and J. K. Barton, *Biochemistry*, 1999, **38**, 4655-4662.

160.   T. Giraldi, Zassinov.G and G. Mestroni, *Chemico-Biological Interactions*, 1974, **9**, 389-394.

161.   G. Sava, S. Zorzet, S. Pacor, G. Mestroni and G. Zassinovich, *Cancer Chemotherapy and Pharmacology*, 1989, **24**, 302-306.

162.   G. Sava, S. Pacor, V. Ceschia, G. Zassinovich and G. Mestroni, *Anticancer Research*, 1989, **9**, 787-790.

163.   G. Sava, T. Giraldi, G. Mestroni and G. Zassinovich, *Chemico-Biological Interactions*, 1983, **45**, 1-6.

164.   G. Craciunescu, V. Scarcia, A. Furlani, E. P. Iglesias, C. Ghirvu and A. Papaioannou, *Anticancer Research*, 1989, **9**, 781-785.

165.   J. Reedijk, *Platinum Metals Review*, 2008, **52**, 2-11.

166.   L. Zhang, P. Carroll and E. Meggers, *Organic Letters*, 2004, **6**, 521-523.

167.   A. Wilbuer, D. H. Vlecken, D. J. Schmitz, K. Kraling, K. Harms, C. P. Bagowski and E. Meggers, *Angewandte Chemie-International Edition*, 2010, **49**, 3839-3842.

168.   Z. Liu and P. J. Sadler, *Accounts of Chemical Research*, 2014, **47**, 1174-1185.

169.   Z. Liu, A. Habtemariam, A. M. Pizarro, G. J. Clarkson and P. J. Sadler, *Organometallics*, 2011, **30**, 4702-4710.

170.   Z. Almodares, S. J. Lucas, B. D. Crossley, A. M. Basri, C. M. Pask, A. J. Hebden, R. M. Phillips and P. C. McGowan, *Inorganic Chemistry*, 2014, **53**, 727-736.

171.   S. J. Lucas, R. M. Lord, R. L. Wilson, R. M. Phillips, V. Sridharan and P. C. McGowan, *Dalton Transactions*, 2012, **41**, 13800-13802.

172.   M. A. Scharwitz, I. Ott, Y. Geldmacher, R. Gust and W. S. Sheldrick, *Journal of Organometallic Chemistry*, 2008, **693**, 2299-2309.

173.   C. R. Munteanu and K. Suntharalingam, *Dalton Transactions*, 2015, **44**, 13796-13808.

174.   R. D. Wright and A. Shulman, *Nature*, 1957, **179**, 425-426.

175.   F. P. Dwyer, I. K. Reid, A. Shulman, G. M. Laycock and S. Dixson, *Australian Journal of Experimental Biology and Medical Science*, 1969, **47**, 203-218.

176.   P. A. Asbell, S. P. Epstein, J. A. Wallace, D. Epstein, C. C. Stewart and R. M. Burger, *Cornea*, 1998, **17**, 550-557.

177.   S. P. Epstein, Y. Y. Pashinsky, D. Gershon, I. Winicov, C. Srivilasa, K. J. Kristic and P. A. Asbell, *BMC Ophthalmology*, 2006, **6**.

178.   J. A. Schwartz, E. K. Lium and S. J. Silverstein, *Journal of Virology*, 2001, **75**, 4117-4128.

179.   P. A. Asbell, S. P. Epstein, J. A. Wallace, J. Tobak, C. C. Stewart and R. M. Burger, *Investigative Ophthalmology & Visual Science*, 1996, **37**, 208-208.

180.   A. Y. Louie and T. J. Meade, *Proceedings of the National Academy of Sciences of the United States of America*, 1998, **95**, 6663-6668.

181.   T. Takeuchi, A. Bottcher, C. M. Quezada, T. J. Meade and H. B. Gray, *Bioorganic & Medicinal Chemistry*, 1999, **7**, 815-819.

182.   A. S. Harney, J. Lee, L. M. Manus, P. J. Wang, D. M. Ballweg, C. LaBonne and T. J. Meade, *Proceedings of the National Academy of Sciences of the United States of America*, 2009, **106**, 13667-13672.

183.   A. S. Harney, T. J. Meade and C. LaBonne, *Plos One*, 2012, **7**.

184.   M. C. Heffern, J. W. Kurutz and T. J. Meade, *Chem.-Eur. J.*, 2013, **19**, 17043-17053.

185.   R. R. Hurtado, A. S. Harney, M. C. Heffern, R. J. Holbrook, R. A. Holmgren and T. J. Meade, *Molecular Pharmaceutics*, 2012, **9**, 325-333.

186.   L. F. Vistain, N. Yamamoto, R. Rathore, P. Cha and T. J. Meade, *Chembiochem*, 2015, **16**, 2065-2072.

187.   A. S. Harney, L. B. Sole and T. J. Meade, *Journal of Biological Inorganic Chemistry*, 2012, **17**, 853-860.

188.   S. P. Osinsky, I. Y. Levitin, A. L. Sigan, L. N. Bubnovskaya, I. I. Ganusevich, L. Campanella and P. Wardman, *Russian Chemical Bulletin International Edition*, 2003, **52**, 2636-2645.

189.   K. Cheng, Q. Z. Zheng and H. L. Zhu, *Inorganic Chemistry Communications*, 2009, **12**, 1116-1119.

190.   K. Dhahagani, S. M. Kumar, G. Chakkaravarthi, K. Anitha, J. Rajesh, A. Ramu and G. Rajagopal, *Spectrochimica Acta Part a-Molecular and Biomolecular Spectroscopy*, 2014, **117**, 87-94.

191.   S. J. Kirubavathy, R. Velmurugan, R. Karvembu, N. S. P. Bhuvanesh, K. Parameswari and S. Chitra, *Russian Journal of Coordination Chemistry*, 2015, **41**, 345-352.

192.   P. Ghosh, A. R. Chowdhury, S. K. Saha, M. Ghosh, M. Pal, N. C. Murmu and P. Banerjee, *Inorganica Chimica Acta*, 2015, **429**, 99-108.

193.   P. Martinez-Bulit, A. Garza-Ortiz, E. Mijangos, L. Barron-Sosa, F. Sanchez-Bartez, I. Gracia-Mora, A. Flores-Parra, R. Contreras, J. Reedijk and N. Barba-Behrens, *Journal of Inorganic Biochemistry*, 2015, **142**, 1-7.

194.   L. Shi, R. Q. Fang, J. Y. Xue, Z. P. Xiao, S. H. Tan and H. L. Zhu, *Australian Journal of Chemistry*, 2008, **61**, 288-296.

195.   D. S. Raja, N. S. P. Bhuvanesh and K. Natarajan, *Dalton Transactions*, 2012, **41**, 4365-4377.

196.   A. Kareem, H. Zafar, A. Sherwani, O. Mohammad and T. A. Khan, *Journal of Molecular Structure*, 2014, **1075**, 17-25.

197.   C. L. Chen, X. F. Zhu, M. X. Li, H. M. Guo and J. Y. Niu, *Russian Journal of Coordination Chemistry*, 2011, **37**, 435-438.

198.   L. F. Chin, S. M. Kong, H. L. Seng, K. S. Khoo, R. Vikneswaran, S. G. Teoh, M. Ahmad, S. B. A. Khoo, M. J. Maah and C. H. Ng, *Journal of Inorganic Biochemistry*, 2011, **105**, 339-347.

199.   D. Dey, A. B. Roy, A. Ranjani, L. Gayathri, S. Chandraleka, D. Dhanasekaran, M. A. Akbarsha, C. Y. Shen, H. L. Tsai, M. Maji, N. Kole and B. Biswas, *Journal of Chemical Sciences*, 2015, **127**, 649-661.

200.   S. Jagadeesan, V. Balasubramanian, P. Baumann, M. Neuburger, D. Haeussinger and C. G. Palivan, *Inorganic Chemistry*, 2013, **52**, 12535-12544.

201.   D. Lahiri, T. Bhowmick, B. Banik, R. Railkar, S. Ramakumar, R. R. Dighe and A. R. Chakravarty, *Polyhedron*, 2010, **29**, 2417-2425.

202.   D. Lahiri, S. Roy, S. Saha, R. Majumdar, R. R. Dighe and A. R. Chakravarty, *Dalton Transactions*, 2010, **39**, 1807-1816.

203.   M. Ahmad, M. Afzal, S. Tabassum, B. Kalniska, J. Mrozinski and P. K. Bharadwaj, *European Journal of Medicinal Chemistry*, 2014, **74**, 683-693.

204.   R. S. Kumar and S. Arunachalam, *Biophysical Chemistry*, 2008, **136**, 136-144.

205.   R. S. Kumar, S. Arunachalam, V. S. Periasamy, C. P. Preethy, A. Riyasdeen and M. A. Akbarsha, *Journal of Inorganic Biochemistry*, 2009, **103**, 117-127.

206.   R. S. Kumar, A. Riyasdeen, M. Dinesh, C. P. Paul, S. Srinag, H. Krishnamurthy, S. Arunachalam and M. A. Akbarsha, *Archiv Der Pharmazie*, 2011, **344**, 422-430.

207.   I. H. Hall, C. B. Lackey, T. D. Kistler, R. W. Durham, E. M. Jouad, M. Khan, X. D. Thanh, S. Djebbar-Sid, O. Benali-Baitich and G. M. Bouet, *Pharmazie*, 2000, **55**, 937-941.

208.   I. H. Hall, K. G. Rajendran, D. X. West and A. E. Liberta, *Anti-Cancer Drugs*, 1993, **4**, 231-240.

209.   R. Manikandan, P. Viswanathamurthi, K. Velmurugan, R. Nandhakumar, T. Hashimoto and A. Endo, *Journal of Photochemistry and Photobiology B-Biology*, 2014, **130**, 205-216.

210.   E. Ramachandran, S. P. Thomas, P. Poornima, P. Kalaivani, R. Prabhakaran, V. V. Padma and K. Natarajan, *European Journal of Medicinal Chemistry*, 2012, **50**, 405-415.

211.   A. M. Downward, R. T. Jane, M. I. J. Polson, E. G. Moore and R. M. Hartshorn, *Dalton Transactions*, 2012, **41**, 14425-14432.

212.   V. Thamilarasan, A. Jayamani and N. Sengottuvelan, *European Journal of Medicinal Chemistry*, 2015, **89**, 266-278.

213.   J. L. Li, L. Jiang, B. W. Wang, J. L. Tian, W. Gu, X. Liu and S. P. Yan, *Journal of Inorganic Biochemistry*, 2015, **145**, 19-29.

214.   I. Tsitsa, A. Tarushi, P. Doukoume, F. Perdih, A. de Almeida, A. Papadopoulos, S. Kalogiannis, A. Casini, I. Turel and G. Psomas, *RSC Advances*, 2016, **6**, 19555-19570.

215.   I. H. Hall, C. B. Lackey, T. D. Kistler, R. W. Durham, J. M. Russell and R. N. Grimes, *Anticancer Research*, 2000, **20**, 2345-2354.

216.   N. Singh, S. Jang, J. H. Jo, D. H. Kim, D. W. Park, I. Kim, H. Kim, S. C. Kang and K. W. Chi, *Chem.-Eur. J.*, 2016, **22**, 16157-16164.

217.   M. C. Heffern, N. Yamamoto, R. J. Holbrook, A. L. Eckermann and T. J. Meade, *Current Opinion in Chemical Biology*, 2013, **17**, 189-196.

218.   D. Osella, M. Ravera, C. Nervi, G. Cavigiolio, M. Vincenti, A. Vessieres and G. Jaouen, *European Journal of Inorganic Chemistry*, 2000, 491-497.

219.   M. Jung, D. E. Kerr and P. D. Senter, *Archiv Der Pharmazie*, 1997, **330**, 173-176.

220.   I. Ott, B. Kircher and R. Gust, *Journal of Inorganic Biochemistry*, 2004, **98**, 485-489.

221.   I. Ott, K. Schmidt, B. Kircher, P. Schumacher, T. Wiglenda and R. Gust, *Journal of Medicinal Chemistry*, 2005, **48**, 622-629.

222.   K. Schmidt, M. Jung, R. Keilitz, B. Schnurr and R. Gust, *Inorganica Chimica Acta*, 2000, **306**, 6-16.

223.   J. R. Vane, *Nature-New Biology*, 1971, **231**, 232-235.

224.   I. Ott, B. Kircher, C. P. Bagowski, D. H. W. Vlecken, E. B. Ott, J. Will, K. Bensdorf, W. S. Sheldrick and R. Gust, *Angewandte Chemie-International Edition*, 2009, **48**, 1160-1163.

225.   I. Zanellato, I. Bonarrigo, M. Ravera, E. Gabano, R. Gust and D. Osella, *Metallomics*, 2013, **5**, 1604-1613.

226.   G. Rubner, K. Bensdorf, A. Wellner, S. Bergemann, I. Ott and R. Gust, *European Journal of Medicinal Chemistry*, 2010, **45**, 5157-5163.

227.   F. Dimiza, A. N. Papadopoulos, V. Tangoulis, V. Psycharis, C. P. Raptopoulou, D. P. Kessissoglou and G. Psomas, *Dalton Transactions*, 2010, **39**, 4517-4528.

228.   D. Kovala-Demertzi, D. Hadjipavlou-Litina, M. Staninska, A. Primikyri, C. Kotoglou and M. A. Demertzis, *Journal of Enzyme Inhibition and Medicinal Chemistry*, 2009, **24**, 742-752.

229.   R. Smolkova, V. Zelenak, L. Smolko and M. Dusek, *Zeitschrift Fur Kristallographie-Crystalline Materials*, 2016, **231**, 715-724.

230.   S. Tsiliou, L. A. Kefala, A. G. Hatzidimitriou, D. P. Kessissoglou, F. Perdih, A. N. Papadopoulos, I. Turel and G. Psomas, *Journal of Inorganic Biochemistry*, 2016, **160**, 125-139.

231.   A. Tarushi, S. Perontsis, A. G. Hatzidimitriou, A. N. Papadopoulos, D. P. Kessissoglou and G. Psomas, *Journal of Inorganic Biochemistry*, 2015, **149**, 68-79.

232.   S. Tsiliou, L. A. Kefala, F. Perdih, I. Turel, D. P. Kessissoglou and G. Psomas, *European Journal of Medicinal Chemistry*, 2012, **48**, 132-142.

233.   P. B. Cressey, A. Eskandari, P. M. Bruno, C. X. Lu, M. T. Hemann and K. Suntharalingam, *Chembiochem*, 2016, **17**, 1713-1718.

234.   F. Dimiza, A. N. Papadopoulos, V. Tangoulis, V. Psycharis, C. P. Raptopoulou, D. P. Kessissoglou and G. Psomas, *Journal of Inorganic Biochemistry*, 2012, **107**, 54-64.

235.   S. Defazio and R. Cini, *Journal of the Chemical Society-Dalton Transactions*, 2002, 1888-1897.

236.   T. H. Sanatkar, H. Hadadzadeh, J. Simpson and Z. Jannesari, *Journal of Molecular Structure*, 2013, **1049**, 336-344.

237.   S. Caglar, I. E. Aydemir, M. Cankaya, M. Kuzucu, E. Temel and O. Buyukgungor, *Journal of Coordination Chemistry*, 2014, **67**, 969-985.

238.   K. Nikitin, Y. Ortin, H. Muller-Bunz, M. A. Plamont, G. Jaouen, A. Vessieres and
        M. J. McGlinchey, *Journal of Organometallic Chemistry*, 2010, **695**, 595-608.

239.   I. Ott, T. Koch, H. Shorafa, Z. L. Bai, D. Poeckel, D. Steinhilber and R. Gust, *Org.
        Biomol. Chem.*, 2005, **3**, 2282-2286.

240.   C. D. Sergeant, I. Ott, A. Sniady, S. Meneni, R. Gust, A. L. Rheingold and R.
        Dembinski, *Org. Biomol. Chem.*, 2008, **6**, 73-80.

241.   M. Schlenk, I. Ott and R. Gust, *Journal of Medicinal Chemistry*, 2008, **51**, 7318-
        7322.

242.   A. Gross, M. Neukamm and N. Metzler-Nolte, *Dalton Transactions*, 2011, **40**,
        1382-1386.

243.   M. A. Neukamm, A. Pinto and N. Metzler-Nolte, *Chemical Communications*,
        2008, 232-234.

244.   A. J. Atkin, S. Williams, P. Sawle, R. Motterlini, J. M. Lynam and I. J. S. Fairlamb,
        Dalton Transactions, 2009, 3653-3656.

245.   Y. G. Gong, T. F. Zhang, M. Li, N. Xi, Y. W. Zheng, Q. Y. Zhao, Y. L. Chen and B.
        Liu, *Free Radical Biology and Medicine*, 2016, **97**, 362-374.

246.   Y. G. Gong, T. F. Zhang, H. P. Liu, Y. W. Zheng, N. Li, Q. Y. Zhao, Y. L. Chen and
        B. Liu, *Transition Metal Chemistry*, 2015, **40**, 413-426.

247.   J. B. Zawilska, J. Wojcieszak and A. B. Olejniczak, *Pharmacological Reports*,
        2013, **65**, 1-14.

248.   J. M. Brown and W. R. William, *Nature Reviews Cancer*, 2004, **4**, 437-447.

249.   G. Ilangovan, H. Q. Li, J. L. Zweier and P. Kuppusamy, *Molecular and Cellular
        Biochemistry*, 2002, **234**, 393-398.

250.   P. Swietach, R. D. Vaughan-Jones, A. L. Harris and A. Hulikova, *Philosophical
        Transactions of the Royal Society B-Biological Sciences*, 2014, **369**.

251.   K. M. Comerford, T. J. Wallace, J. Karhausen, N. A. Louis, M. C. Montalto and
        S. P. Colgan, *Cancer Research*, 2002, **62**, 3387-3394.

252.   W. A. Denny, *European Journal of Medicinal Chemistry*, 2001, **36**, 577-595.

253.   N. Graf and S. J. Lippard, *Advanced Drug Delivery Reviews*, 2012, **64**, 993-
        1004.

254.   T. W. Hambley, *Dalton Transactions*, 2007, 4929-4937.

255.   Y. Chen and L. Q. Hu, *Medicinal Research Reviews*, 2009, **29**, 29-64.

256.   W. A. Denny, *Cancer Investigation*, 2004, **22**, 604-619.

257.   H. Ostergaard, C. Tachibana and J. R. Winther, *Journal of Cell Biology*, 2004, **166**, 337-345.

258.   F. Q. Schafer and G. R. Buettner, *Free Radical Biology and Medicine*, 2001, **30**, 1191-1212.

259.   L. C. Perrin, W. R. Wilson, W. A. Denny and W. D. McFadyen, *Anti-Cancer Drug Design*, 1999, **14**, 231-241.

260.   P. Brookes and P. D. Lawley, *Biochemical Journal*, 1961, **80**, 496-503.

261.   L. F. Povirk and D. E. Shuker, *Mutation Research-Reviews in Genetic Toxicology*, 1994, **318**, 205-226.

262.   B. A. Teicher, J. L. Jacobs, K. N. S. Cathcart, M. J. Abrams, J. F. Vollano and D. H. Picker, *Radiation Research*, 1987, **109**, 36-46.

263.   B. A. Teicher, M. J. Abrams, K. W. Rosbe and T. S. Herman, *Cancer Research*, 1990, **50**, 6971-6975.

264.   D. C. Ware, P. J. Brothers, G. R. Clark, W. A. Denny, B. D. Palmer and W. R. Wilson, *Journal of the Chemical Society-Dalton Transactions*, 2000, 925-932.

265.   D. C. Ware, B. G. Siim, K. G. Robinson, W. A. Denny, P. J. Brothers and G. R. Clark, *Inorganic Chemistry*, 1991, **30**, 3750-3757.

266.   D. C. Ware, W. A. Denny and G. R. Clark, *Acta Crystallographica Section C-Crystal Structure Communications*, 1997, **53**, 1058-1059.

267.   D. C. Ware, B. D. Palmer, W. R. Wilson and W. A. Denny, *Journal of Medicinal Chemistry*, 1993, **36**, 1839-1846.

268.   D. C. Ware, W. R. Wilson, W. A. Denny and C. E. F. Rickard, *Journal of the Chemical Society-Chemical Communications*, 1991, 1171-1173.

269.   R. F. Anderson, W. A. Denny, D. C. Ware and W. R. Wilson, *British Journal of Cancer*, 1996, **74**, S48-S51.

270.   D. C. Ware, H. R. Palmer, P. J. Brothers, C. E. F. Rickard, W. R. Wilson and W. A. Denny, *Journal of Inorganic Biochemistry*, 1997, **68**, 215-224.

271.   D. C. Ware, H. R. Palmer, F. B. Pruijn, R. E. Anderson, P. J. Brothers, W. A. Denny and W. R. Wilson, *Anti-Cancer Drug Design*, 1998, **13**, 81-103.

272.   P. R. Craig, P. J. Brothers, G. R. Clark, W. R. Wilson, W. A. Denny and D. C. Ware, *Dalton Transactions*, 2004, 611-618.

273.   I. C. A. de Souza, L. V. Faro, C. B. Pinheiro, D. T. G. Gonzaga, F. D. da Silva, V. F. Ferreira, F. D. Miranda, M. Scarpellini and M. Lanznaster, *Dalton Transactions*, 2016, **45**, 13671-13674.

274.   D. Tomco, S. Schmitt, B. Ksebati, M. J. Heeg, Q. P. Dou and C. N. Verani, *Journal of Inorganic Biochemistry*, 2011, **105**, 1759-1766.

275.   D. Tomco, F. R. Xavier, M. M. Allard and C. N. Verani, *Inorganica Chimica Acta*, 2012, **393**, 269-275.

276.   A. Merdad, S. Karim, H. J. Schulten, A. Dallol, A. Buhmeida, F. Al-Thubaity, M. A. Gari, A. G. A. Chaudhary, A. M. Abuzenadah and M. H. Al-Qahtani, *Anticancer Research*, 2014, **34**, 1355-1366.

277.   S. R. Bramhall, M. T. Hallissey, J. Whiting, J. Scholefield, G. Tierney, R. C. Stuart, R. E. Hawkins, P. McCulloch, T. Maughan, P. D. Brown and M. Baillet, *British Journal of Cancer*, 2002, **86**, 1864-1870.

278.   T. W. Failes, C. Cullinane, C. I. Diakos, N. Yamamoto, J. G. Lyons and T. W. Hambley, *Chem.-Eur. J.*, 2007, **13**, 2974-2982.

279.   T. W. Failes and T. W. Hambley, *Dalton Transactions*, 2006, 1895-1901.

280.   P. D. Bonnitcha, B. J. Kim, R. K. Hocking, J. K. Clegg, P. Turner, S. M. Neville and T. W. Hambley, *Dalton Transactions*, 2012, **41**, 11293-11304.

281.   N. Yamamoto, A. K. Renfrew, B. J. Kim, N. S. Bryce and T. W. Hambley, *Journal of Medicinal Chemistry*, 2012, **55**, 11013-11021.

282.   G. O. Ahn, K. J. Botting, A. V. Patterson, D. C. Ware, M. Tercel and W. R. Wilson, *Biochemical Pharmacology*, 2006, **71**, 1683-1694.

283.   G. O. Ahn, D. C. Ware, W. A. Denny and W. R. Wilson, *Radiation Research*, 2004, **162**, 315-325.

284.   J. Y. C. Chang, R. J. Stevenson, G. L. Lu, P. J. Brothers, G. R. Clark, W. A. Denny and D. C. Ware, *Dalton Transactions*, 2010, **39**, 11535-11550.

285.   G. L. Lu, R. J. Stevenson, J. Y. C. Chang, P. J. Brothers, D. C. Ware, W. R. Wilson, W. A. Denny and M. Tercel, *Bioorganic & Medicinal Chemistry*, 2011, **19**, 4861-4867.

286.   J. B. J. Milbank, R. J. Stevenson, D. C. Ware, J. Y. C. Chang, M. Tercel, G. O. Ahn, W. R. Wilson and W. A. Denny, *Journal of Medicinal Chemistry*, 2009, **52**, 6822-6834.

287.   J. Y. C. Chang, G. L. Lu, R. J. Stevenson, P. J. Brothers, G. R. Clark, K. J. Botting, D. M. Ferry, M. Tercel, W. R. Wilson, W. A. Denny and D. C. Ware, *Inorganic Chemistry*, 2013, **52**, 7688-7698.

288.   S. Banerjee and A. R. Chakravarty, *Accounts of Chemical Research*, 2015, **48**, 2075-2083.

289. A. Garai, I. Pant, S. Banerjee, B. Banik, P. Kondaiah and A. R. Chakravarty, *Inorganic Chemistry*, 2016, **55**, 6027-6035.

290. A. K. Renfrew, N. S. Bryce and T. Hambley, *Chem.-Eur. J.*, 2015, **21**, 15224-15234.

291. A. K. Renfrew, N. S. Bryce and T. W. Hambley, *Chemical Science*, 2013, **4**, 3731-3739.

292. T. Sarkar, S. Banerjee and A. Hussain, *RSC Advances*, 2015, **5**, 16641-16653.

293. A. K. Petrus, T. J. Fairchild and R. P. Doyle, *Angewandte Chemie-International Edition*, 2009, **48**, 1022-1028.

294. J. D. Bagnato, A. L. Eilers, R. A. Horton and C. B. Grissom, *Journal of Organic Chemistry*, 2004, **69**, 8987-8996.

295. A. H. Bayomi, A. M. Mitchell, E. N. Natarajan, C. B. Grissom, F. G. West, M. A. Aziza and O. ElAhmady, *Abstracts of Papers of the American Chemical Society*, 1997, **213**, 123-MEDI.

296. S. Mundwiler, B. Spingler, P. Kurz, S. Kunze and R. Alberto, *Chem.-Eur. J.*, 2005, **11**, 4089-4095.

297. P. Ruiz-Sanchez, C. Konig, S. Ferrari and R. Alberto, *Journal of Biological Inorganic Chemistry*, 2011, **16**, 33-44.

298. P. Siega, J. Wuerges, F. Arena, E. Gianolio, S. N. Fedosov, R. Dreos, S. Geremia, S. Aime and L. Randaccio, *Chem.-Eur. J.*, 2009, **15**, 7980-7989.

299. R. Waibel, H. Treichler, N. G. Schaefer, D. R. van Staveren, S. Mundwiler, S. Kunze, M. Kuenzi, R. Alberto, J. Nuesch, A. Knuth, H. Moch, R. Schibli and P. A. Schubiger, *Cancer Research*, 2008, **68**, 2904-2911.

300. N. Viola-Villegas, A. E. Rabideau, M. Bartholoma, J. Zubieta and R. P. Doyle, *Journal of Medicinal Chemistry*, 2009, **52**, 5253-5261.

301. T. A. Shell and D. S. Lawrence, *Accounts of Chemical Research*, 2015, **48**, 2866-2874.

302. J. A. Bauer, *Anti-Cancer Drugs*, 1998, **9**, 239-244.

303. J. A. Bauer, B. H. Morrison, R. W. Grane, B. S. Jacobs, S. Dabney, A. M. Gamero, K. A. Carnevale, D. J. Smith, J. Drazba, B. Seetharam and D. J. Lindner, *Journal of the National Cancer Institute*, 2002, **94**, 1010-1019.

304. J. A. Bauer, G. Frye, A. Bahr, J. Gieg and P. Brofman, *Investigational new drugs*, 2010, **28**, 694-702.

305.   R. L. Chapman, F. S. Stephens and R. S. Vagg, *Inorganica Chimica Acta-Articles*, 1981, **52**, 161-168.

306.   G. Condorelli, A. Seminara and A. Musumeci, *Journal of Inorganic & Nuclear Chemistry*, 1974, **36**, 3763-3766.

307.   K. Nonoyama, *Journal of Inorganic & Nuclear Chemistry*, 1975, **37**, 2449-2453.

308.   M. Ray, R. Mukherjee, J. F. Richardson and R. M. Buchanan, *Journal of the Chemical Society-Dalton Transactions*, 1993, 2451-2457.

309.   G. V. Tsintsadze, R. A. Kiguradze, A. N. Shnulin and K. S. Mamedov, *Journal of Structural Chemistry*, 1986, **27**, 272-278.

310.   S. H. van Rijt, A. J. Hebden, T. Amaresekera, R. J. Deeth, G. J. Clarkson, S. Parsons, P. C. McGowan and P. J. Sadler, *Journal of Medicinal Chemistry*, 2009, **52**, 7753-7764.

311.   Z. Q. Xu, L. K. Thompson, D. A. Black, C. Ralph, D. O. Miller, M. A. Leech and J. A. K. Howard, *Journal of the Chemical Society-Dalton Transactions*, 2001, 2042-2048.

312.   A. M. Basri, R. M. Lord, S. J. Allison, A. Rodriguez-Barzano, S. J. Lucas, F. D. Janeway, H. J. Shepherd, C. M. Pask, R. M. Phillips and P. C. McGowan, *Chem.-Eur. J.*, 2017, **23**, 6341-6356.

313.   R. M. Lord, S. M. Lord, C. M. Pask and P. C. McGowan, *Polyhedron*, 2016, **116**, 136-143.

314.   C. Sambiagio, R. H. Munday, S. P. Marsden, A. J. Blacker and P. C. McGowan, *Chem.-Eur. J.*, 2014, **20**, 17606-17615.

315.   Z. Almodares, PhD Thesis, University of Leeds, 2010.

316.   S. J. Lucas, R. M. Lord, A. M. Basri, S. J. Allison, R. M. Phillips, A. J. Blacker and P. C. McGowan, *Dalton Transactions*, 2016, **45**, 6812-6815.

Chapter 2

Synthesis and characterisation of cobalt (III) *tris*-picolinamide complexes

2. Cobalt (III) *tris*-picolinamide complexes

In comparison to other transition metals, particularly ruthenium, the study of picolinamide ligands with cobalt is in its infancy. At present there are only a limited number of cobalt(III) *tris*-picolinamide complexes which have been published in the literature. The first synthesis of these complexes was reported by Chan and revealed octahedral cobalt(III) complexes with six nitrogen atoms in the inner coordination sphere provided by three bidentate N-aryl picolinamide ligands.[1] These complexes went on to show high activity and selectivity in the catalytic oxidation of ethylbenzene to acetophenone.[1] An alternative synthetic route to these complexes was reported five years later by Gupta with further investigation into the anti-bacterial properties of these complexes, with the complex containing a 2-pyridyl group in particular displaying strong activity against various strains of *Pseudomonas*, *E. coli* and *Shigella*.[2] Complexes containing picolinamide ligands coordinated through the two nitrogen atoms can be converted to an isomer where one of the ligands is instead coordinated through one nitrogen and an oxygen atom. Acidic conditions permit the protonation of one of the ligands which then switches its coordination from *N,N* to *N,O* resulting in a monocationic complex (**Figure 2.1**).[3] In ruthenium picolinamide complexes, *N,N* coordination of the ligand is associated with improved anti-cancer activity compared to *N,O* coordination.[4]
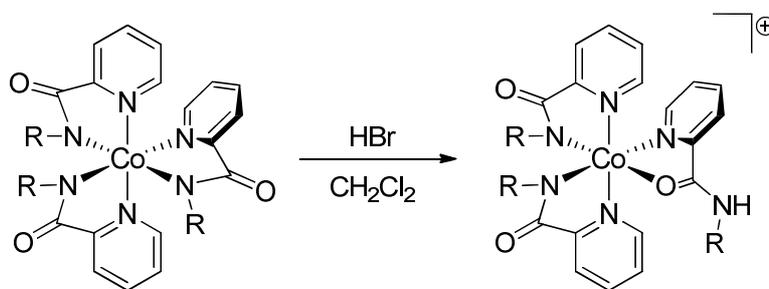
Figure 2.1: Conversion of picolinamide ligand coordination modes

Related cobalt(III) complexes with the same inner coordination sphere provided by two tridentate picolinamide ligands have also been reported.[5] The complex containing a 2-pyridyl group was also found to show promise as an anti-bacterial agent, in common with the analogous bidentate picolinamide complex.[2] When a nitrogen atom is incorporated into the picolinamide aryl ring, this confers the ability to coordinate an additional metal centre resulting in heterobimetallic complexes which can proceed to form networks (**Figure 2.2**). To date, complexes of cobalt(III) with zinc(II), cadmium(II) and mercury(II) have been reported.[6-9]

Bidentate picolinamide ligands may also act as tridentate ligands if the substituent on the aryl ring is a coordinating group such as a thiol.[10]

M = Zn, Cd, Hg
X = Cl, NO₃, solvent
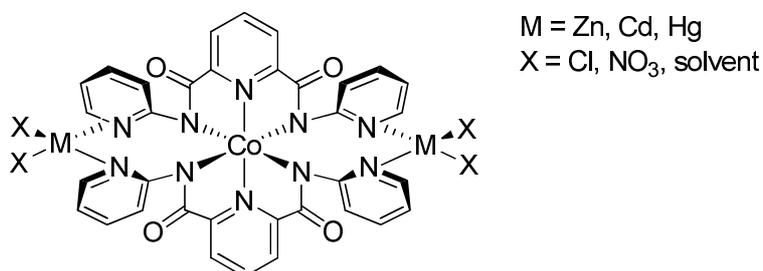
Figure 2.2: Cobalt heterobimetallic complexes reported

2.1 Picolinamide ligands

Picolinamide ligands have the ability to coordinate to a metal centre in a number of different ways: as neutral *N,O* donors or monoanionic *N,N* or *N,O* donors (**Figure 2.3**). Complexes containing more than one picolinamide ligand therefore have the potential to contain ligands with different coordination modes. *N,N* coordination to the metal centre stabilises higher oxidation states due to the strong σ-donor effect, whereas *N,O* coordination tends to stabilise lower oxidation states.[11, 12] The nature of the ligand coordination can thus affect not only the chemical but also the biological properties of the complex as has been observed previously.[4]
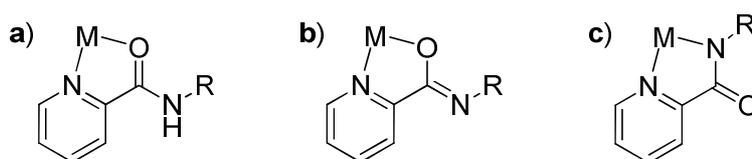
Figure 2.3: Coordination modes for picolinamide ligand a) neutral *N,O* donor b) monoanionic *N,O* donor c) monoanionic *N,N* donor

The N-aryl picolinamide ligands used in this work are shown in **Figure 2.4**. Ligands with electron withdrawing or electron donating groups were synthesised in order to examine the effect of electronics on the properties of the complex. Fluoro-containing ligands were synthesised with the aim of increasing efficacy, as a number of successful drugs contain fluoro groups.[13, 14] All ligands apart from ligand **1.22** have been synthesised previously in the literature and are not discussed further.[15-20] Ligands were synthesised *via* the condensation reaction of picolinic

acid with the respective aniline in the presence of pyridine and triphenylphosphite (**Scheme 2.1a**).[21] Synthesis of ligand **1.11** by this method resulted in poor yields (< 10 %) and was therefore synthesised *via* an alternative condensation reaction involving dichloromethane, triethylamine and phosphorus oxychloride (**Scheme 2.1b**).[22]
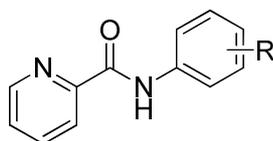
R =	H	(**1.1**)	3-Cl	(**1.7**)	4-CF$_3$	(**1.13**)	3,5-diMe	(**1.19**)
	2-F	(**1.2**)	4-Cl	(**1.8**)	3,5-diCF$_3$	(**1.14**)	4-OMe	(**1.20**)
	3-F	(**1.3**)	3-Br	(**1.9**)	2-Me	(**1.15**)	3,5-diOMe	(**1.21**)
	4-F	(**1.4**)	4-Br	(**1.10**)	3-Me	(**1.16**)	anthracene	(**1.22**)
	2,4-diF	(**1.5**)	2-CF$_3$	(**1.11**)	4-Me	(**1.17**)		
	2,5-diF	(**1.6**)	3-CF$_3$	(**1.12**)	2,3-diMe	(**1.18**)		

Figure 2.4: Picolinamide ligands synthesised within this thesis
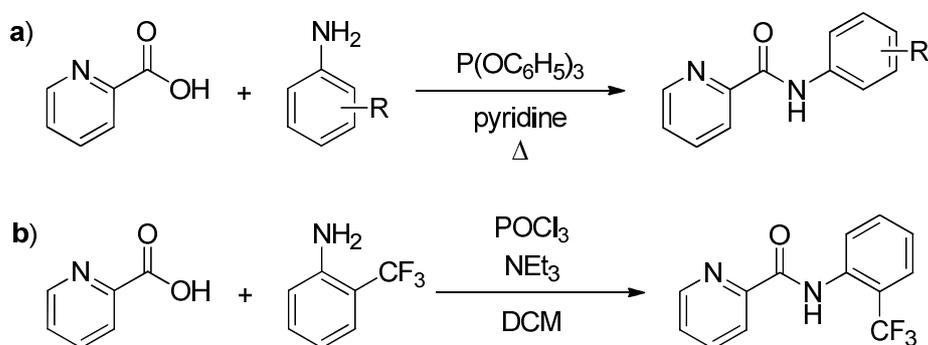
Scheme 2.1: Synthetic routes for picolinamide ligands

The ^{1}H NMR spectra of the picolinamide ligands are all very similar with the pyridyl protons (7.48 - 8.81 ppm) typically having higher chemical shifts than the aryl protons (6.81 - 8.64 ppm). A broad singlet corresponding to the amide proton appears downfield, in the range 9.95 - 10.93 ppm depending upon the nature of the substitution on the aryl ring; this signal is the most characteristic for ligand formation.

2.1.1 X-Ray crystallography of ligand 1.22

Ligand **1.22** crystallised in a monoclinic cell and structural solutions were performed in the $P\,2_1$ space group with a single molecule in the asymmetric unit (**Figure 2.5**). Selected bond lengths and angles are stated in **Table 2.1**. There is evidence within the crystal structure for an intramolecular hydrogen bond between the amide proton and the pyridyl nitrogen (N(1)...N(2) = 2.662(6) Å).
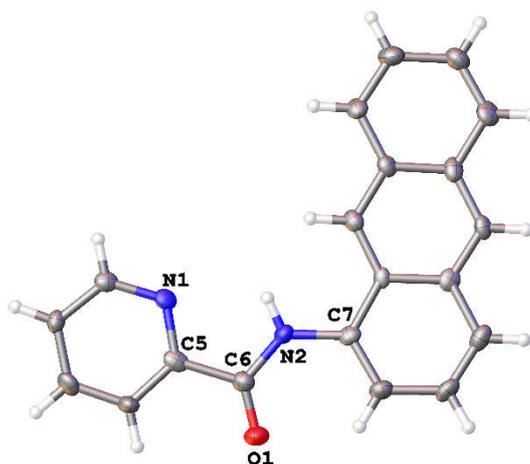
Figure 2.5: Molecular structure of ligand **1.22**. Displacement ellipsoids are at the 50 % probability level

Table 2.1: Selected bond lengths and angles for ligand **1.22**

Bond	Distance / Å	Bond	Angle / °
C(5)-C(6)	1.328(6)	N(1)-C(5)-C(6)	116.9(4)
C(6)-O(1)	1.216(6)	C(5)-C(6)-O(1)	120.6(5)
C(6)-N(2)	1.354(6)	O(1)-C(6)-N(2)	126.2(5)
N(2)-C(7)	1.412(6)	C(6)-N(2)-C(7)	129.3(4)

2.2 Cobalt *tris*-picolinamide complexes (*N,N* coordination)

The cobalt picolinamide complexes synthesised within this section are shown in **Figure 2.6**. Complexes were synthesised *via* the complexation reaction of picolinamide ligand with cobalt chloride in the presence of triethylamine, similar to the synthesis reported by Qi *et al*. (**Scheme 2.2**).[1]
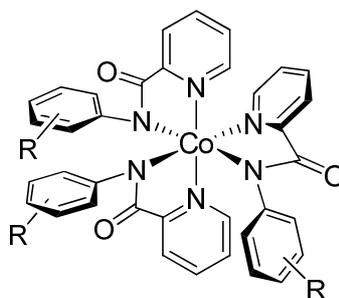
R =	H	(**2.1a**)	2,5-diF	(**2.6**)	3-CF$_3$	(**2.11**)	3,5-diMe	(**2.16a**)
	2-F	(**2.2**)	3-Cl	(**2.7**)	4-CF$_3$	(**2.12**)	4-OMe	(**2.17a**)
	3-F	(**2.3**)	4-Cl	(**2.8**)	3,5-diCF$_3$	(**2.13**)	3,5-diOMe	(**2.18a**)
	4-F	(**2.4**)	3-Br	(**2.9**)	3-Me	(**2.14a**)		
	2,4-diF	(**2.5**)	4-Br	(**2.10**)	4-Me	(**2.15a**)		

Figure 2.6: Cobalt picolinamide complexes synthesised within this section
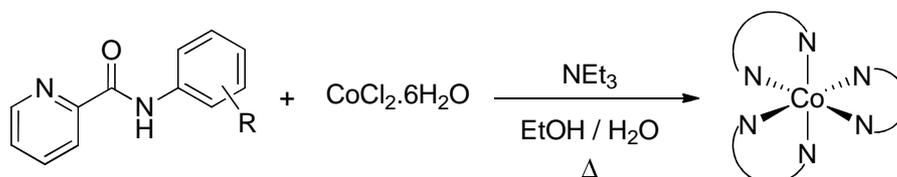
Scheme 2.2: Synthetic route for cobalt *tris*-picolinamide complexes in this section

In this synthesis, cobalt is oxidised from Co(II) in the metal chloride precursor to Co(III) in the metal complex, which then permits the complexation of three ligands to the metal centre. All three ligands coordinate through the pyridyl nitrogen and amide nitrogen, acting as monoanionic donors to help stabilise the 3+ oxidation state of the central cobalt ion, as has been reported previously.[1, 2] The presence of triethylamine as a base in the reaction appears to be responsible for the oxidation of the cobalt in an aerobic atmosphere. This oxidation can also occur with other bases including additional amine containing bases, hydroxides, carbonates and to a lesser extent hydrogen carbonates; this observation is consistent with the literature.[23] Cobalt may also be oxidised with hydrogen peroxide followed by

complexation to the deprotonated ligand, however this reaction results in poorer conversion of the metal precursor to the final complex. The synthesis was successful with the majority of picolinamide ligands, however larger groups in the *ortho* position on the aryl ring were not well tolerated. Complexes with a proton or a fluorine atom at the *ortho* position were successfully synthesised but attempts with larger atoms, such as chlorine, were unsuccessful; this was presumably due to steric crowding around the metal centre.

2.2.1 Characterisation of cobalt *tris*-picolinamide complexes (*N,N* coordination)

The cobalt(III) picolinamide complexes are low spin d^6 and thus diamagnetic. The ^{1}H NMR spectra of these complexes are all very similar with the pyridyl protons (9.24 - 6.98 ppm) typically having higher chemical shifts than the aryl protons (7.87 - 4.99 ppm). Pyridyl and aryl protons tend to be shifted upfield in the complex in comparison to in the free ligand, however one set of pyridyl protons are shifted downfield. Examples of the ^{1}H NMR chemical shifts for complex **2.7** compared to the ligands are shown in **Figure 2.7** and **Table 2.2**. Three distinct or overlapping sets of signals for the picolinamide protons are seen in the final complex as the three ligands are inequivalent due to the slightly distorted octahedral geometry of the complex.
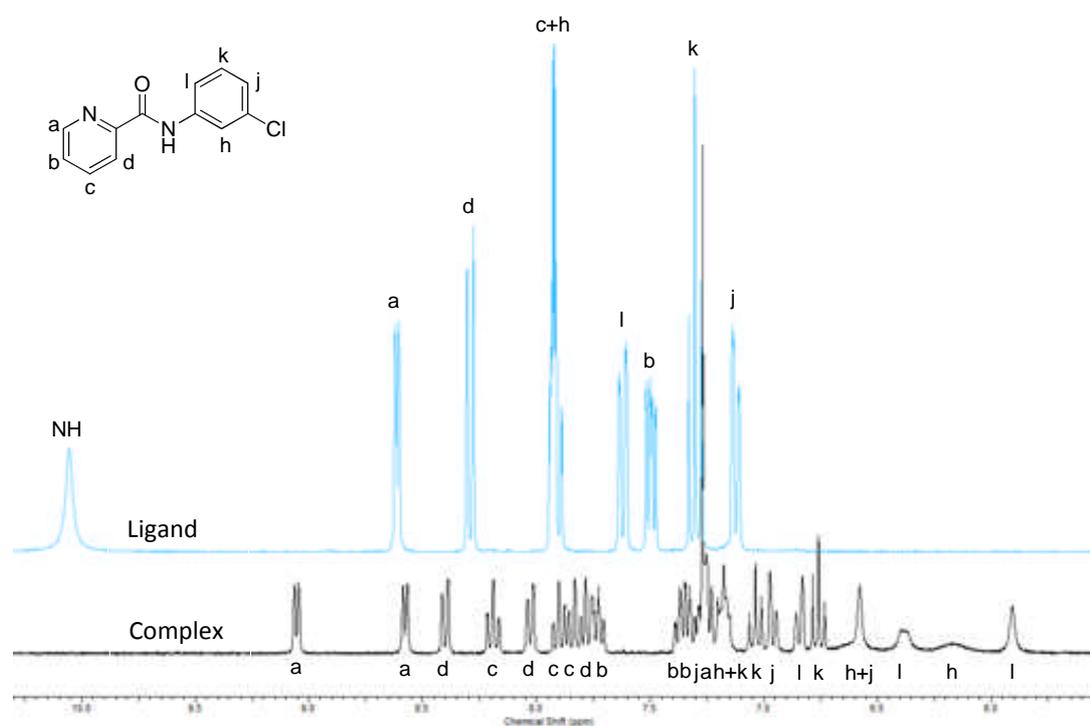
Figure 2.7: ^{1}H NMR spectra of complex **2.7** and ligand **1.7**

Table 2.2: ^{1}H NMR chemical shifts for complex **2.7** and ligand **1.7**

Proton environment	^{1}H NMR chemical shift / ppm	
	Ligand	Complex
Pyridyl	8.63	9.04, 8.57 and 7.23
	8.32	8.38, 8.02 and 7.77
	7.95	8.17, 7.89 and 7.82
	7.52	7.71, 7.36 and 7.31
Aryl	7.95	6.84, 6.37 and 5.89
	7.64	7.17, 6.58 and 6.18
	7.32	7.17, 7.02 and 6.75
	7.14	7.26, 6.95 and 6.58

The IR spectra of complexes **2.1a** - **2.18a** all display a weak aryl CH stretch at 3035 - 3090 cm^{-1} and a two medium intensity CO stretches at 1615 - 1635 cm^{-1} and 1585 - 1600 cm^{-1}. There are also several bands of varying intensities in the region 500 - 1600 cm^{-1}. The disappearance of the broad NH stretches (3250 - 3400 cm^{-1}) is characteristic of complex formation as the picolinamide ligands coordinate through the amide nitrogen. The CO stretch is shifted to a lower wavenumber in the complex compared to the free ligand as is expected due to the weakening of the CO bond following coordination to the metal centre.[1-3]

The UV/vis spectrum of complex **2.1a** is shown in **Figure 2.8**. The spectrum shows an intense ligand based absorbance (π-π*) at approximately 200 nm with two less intense charge-transfer bounds at around 265 and 300 nm. No d-d transitions are seen in the 400 - 800 nm region.[1-3]
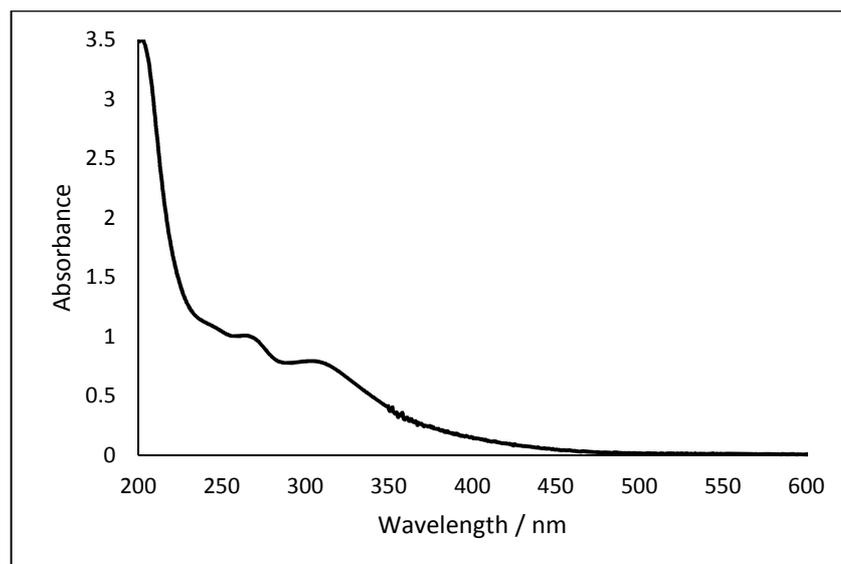
Figure 2.8: UV/vis spectrum of complex **2.1a**

2.2.2 X-Ray crystallography of cobalt *tris*-picolinamide complexes (*N,N* coordination)

Complexes crystallised in either a monoclinic (**2.2**, **2.3**, **2.6**, **2.7**, **2.10**, **2.13**) or triclinic (**2.1a**, **2.4**, **2.5**, **2.8**, **2.9**, **2.11**, **2.12**, **2.14a**, **2.15a** - **2.17a**) cell and structural solutions were performed in the $P\,2_1/n$ (**2.3**, **2.6**), $P\,2_1/c$ (**2.2**, **2.7**, **2.10**, **2.13**) or $P\,\overline{1}$ (**2.1**, **2.4**, **2.5**, **2.8**, **2.9**, **2.11**, **2.12**, **2.14a** - **2.17a**) space group with one (**2.2**, **2.3**, 2.6 - **2.11**, **2.13** - **2.17a**), two (**2.4**, **2.5**, **2.12**, **2.16a**) or three (**2.1a**) molecules in the asymmetric unit (**Figure 2.9**). Selected bond lengths and angles are stated in **Table 2.3** and **Table 2.4** respectively.

2.1a   **2.2**
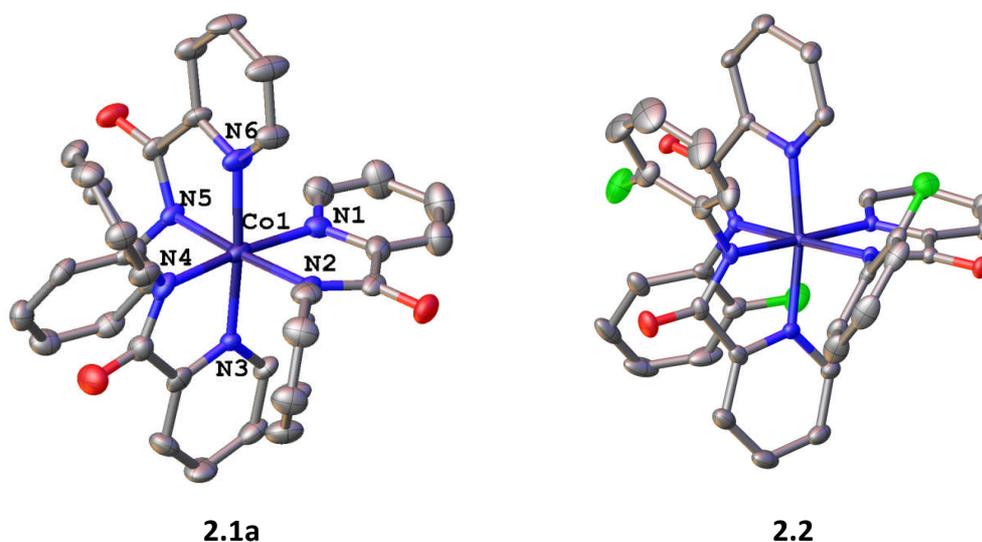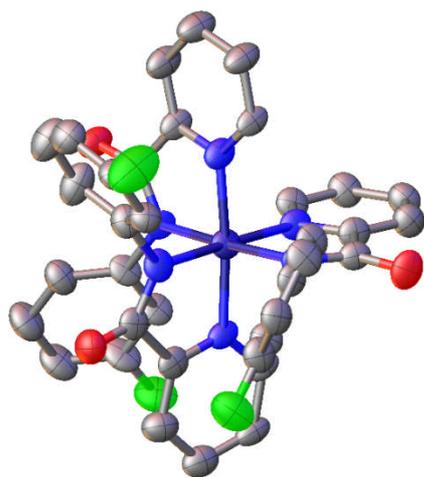
2.3
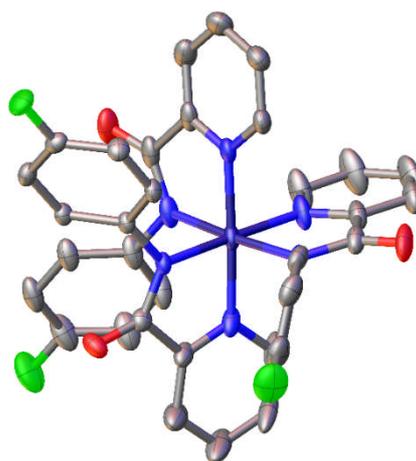
2.4
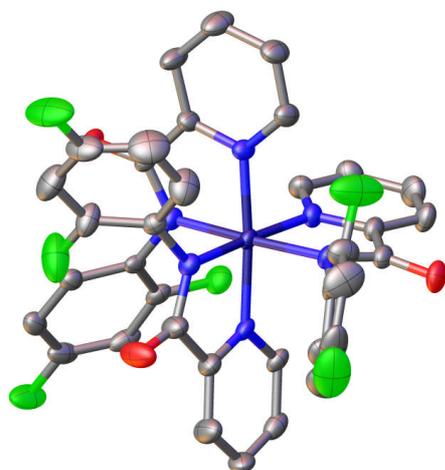
2.5
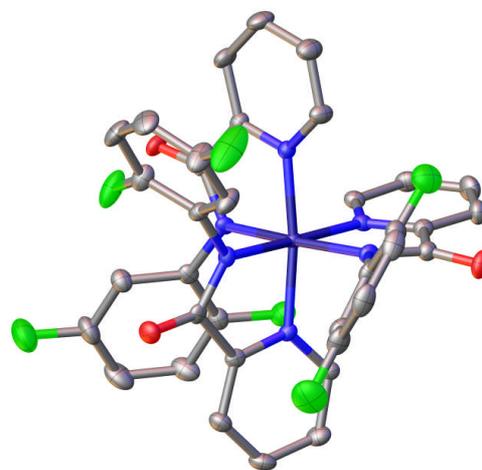
2.6
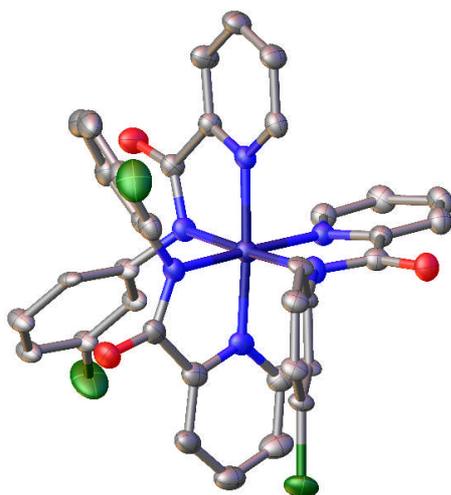
2.7
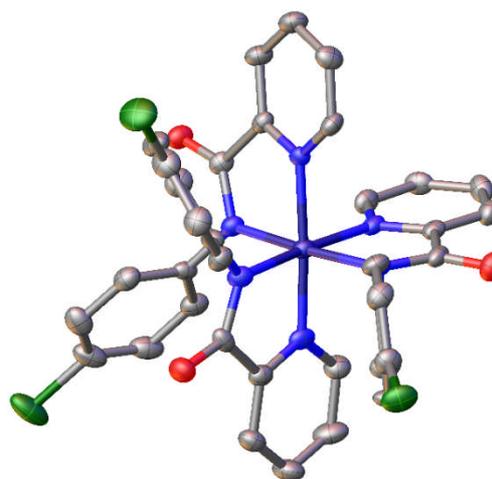
2.8
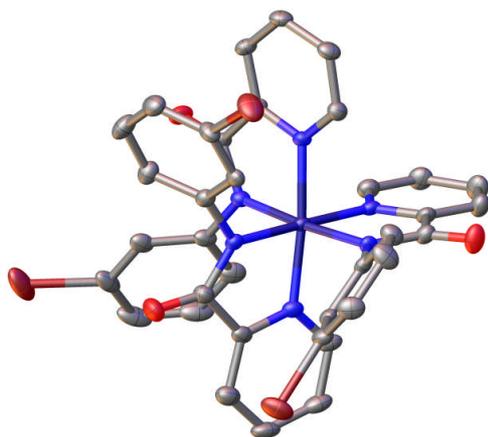
2.9
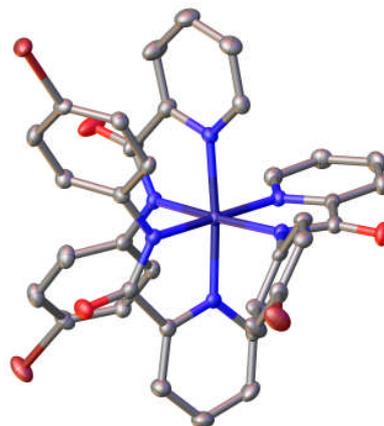
2.10

2.11

2.12

2.13

2.14a

2.15a   **2.16a**
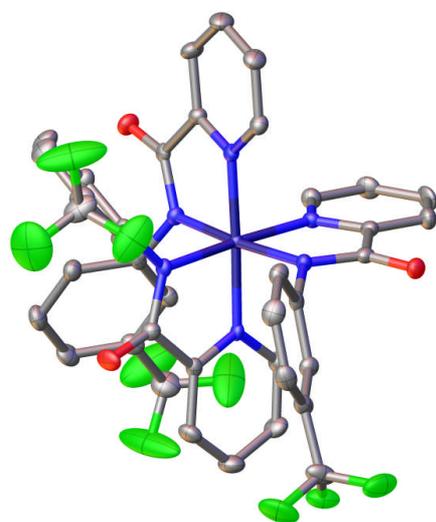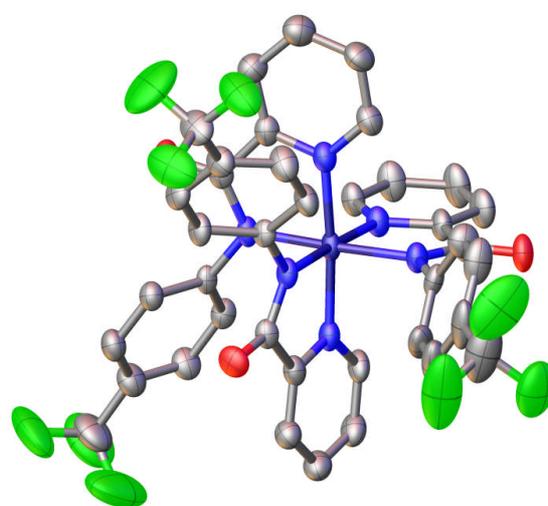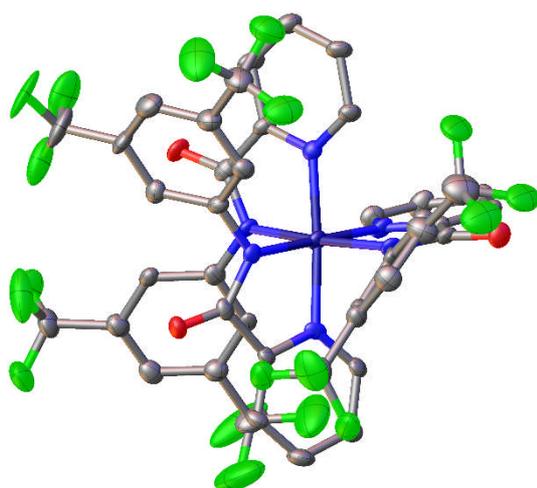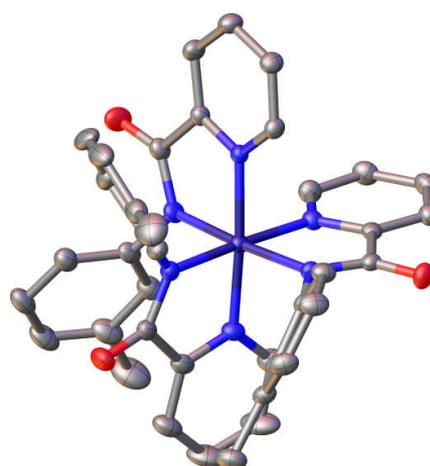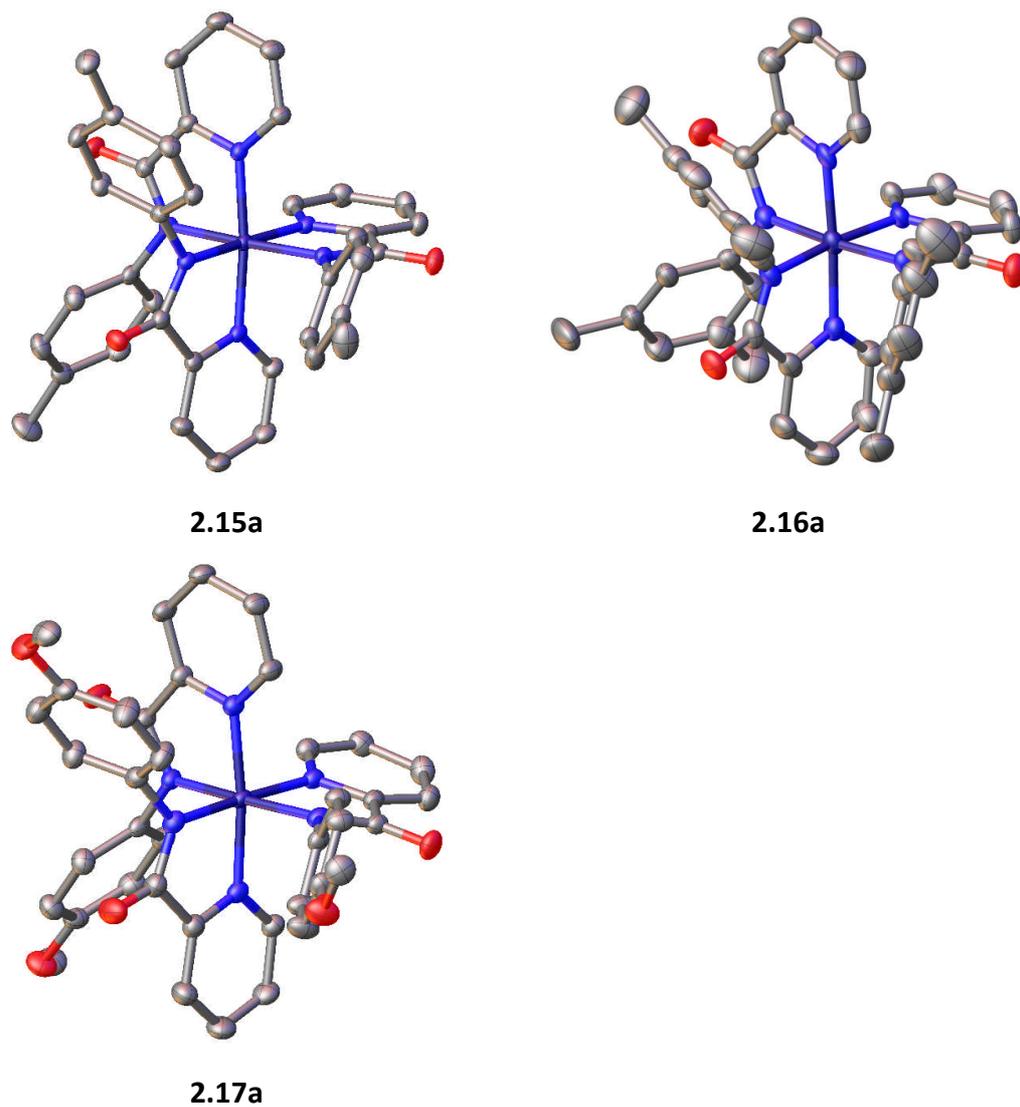
2.17a

Figure 2.9: Molecular structures of complexes **2.1a** - **2.17a**. Displacement ellipsoids are at the 50 % probability level. Hydrogen atoms and co-crystallising solvent molecules are omitted for clarity. For clarity only complex **2.1a** is labelled, the remaining complexes follow the same atom numbering scheme

Table 2.3: Selected bond lengths for complexes **2.1a** - **2.17a**

	Distance / Å					
	Co(1)-N(1)	**Co(1)-N(2)**	**Co(1)-N(3)**	**Co(1)-N(4)**	**Co(1)-N(5)**	**Co(1)-N(6)**
2.1a	1.968(4)	1.962(5)	1.928(5)	1.955(5)	1.961(4)	1.928(5)
2.2	1.9688(18)	1.9738(19)	1.9325(18)	1.9479(19)	1.9512(18)	1.9211(18)
2.3	1.955(9)	1.981(7)	1.950(8)	1.939(9)	1.961(7)	1.949(9)
2.4	1.954(2)	1.968(2)	1.942(2)	1.9411(18)	1.968(2)	1.921(2)
2.5	1.962(4)	1.977(4)	1.936(4)	1.949(4)	1.963(4)	1.913(4)
2.6	1.973(3)	1.968(3)	1.932(3)	1.942(3)	1.965(2)	1.926(3)
2.7	1.966(4)	1.966(4)	1.949(4)	1.933(3)	1.936(4)	1.924(4)
2.8	1.974(5)	1.957(5)	1.952(4)	1.936(5)	1.955(5)	1.918(4)
2.9	1.962(3)	1.974(3)	1.932(3)	1.939(3)	1.949(3)	1.937(3)
2.10	1.965(2)	1.966(2)	1.924(2)	1.942(2)	1.947(2)	1.933(2)
2.11	1.962(3)	1.961(3)	1.933(3)	1.939(3)	1.929(3)	1.919(3)
2.12	1.954(5)	1.969(5)	1.933(6)	1.944(5)	1.951(5)	1.929(6)
2.13	1.954(7)	1.977(7)	1.937(7)	1.950(7)	1.975(7)	1.942(7)
2.14a	1.9567(15)	1.9636(13)	1.9278(15)	1.9336(15)	1.9497(14)	1.9420(14)
2.15a	1.9289(18)	1.9298(19)	1.9323(18)	1.9526(18)	1.9694(19)	1.9720(18)
2.16a	1.958(7)	1.963(6)	1.946(7)	1.945(7)	1.944(7)	1.935(7)
2.17a	1.932(2)	1.944(2)	1.934(2)	1.938(2)	1.960(2)	1.985(2)

Table 2.4: Selected bond angles for complexes **2.1a** - **2.17a**

	Angle / °		
	N(1)-Co(1)-N(2)	**N(2)-Co(1)-N(3)**	**N(2)-Co(1)-N(4)**
2.1a	81.66(18)	89.68(19)	95.4(2)
2.2	81.56(7)	88.99(8)	96.29(8)
2.3	82.5(3)	88.0(3)	95.8(3)
2.4	82.04(9)	88.19(9)	95.93(8)
2.5	81.21(15)	87.74(16)	94.67(15)
2.6	82.03(11)	88.46(11)	96.70(11)
2.7	82.04(15)	87.96(16)	94.66(15)
2.8	82.2(2)	89.24(19)	93.7(2)
2.9	82.43(13)	89.97(13)	96.92(13)
2.10	82.42(9)	87.56(10)	97.27(9)
2.11	82.18(12)	86.77(12)	95.47(13)
2.12	82.1(2)	88.6(2)	94.9(2)
2.13	82.3(3)	87.7(3)	93.2(3)
2.14a	82.37(6)	89.23(6)	95.19(6)
2.15a	81.79(8)	89.16(7)	95.88(8)
2.16a	82.4(3)	88.7(3)	94.4(3)
2.17a	81.49(9)	88.32(8)	95.41(9)

All complexes show a distorted octahedral geometry around the central cobalt atom with the relevant bond angles in the range 81.5 - 97.3°, deviating from the expected 90° (**Table 2.4**). This distortion can be ascribed to the restrictions imposed by the bidentate picolinamide ligand. The cobalt-pyridyl nitrogen bond lengths are in the range 1.92 - 1.99 Å and the cobalt-amide nitrogen bond lengths are in the range 1.93 - 1.98 Å. This is consistent with related low spin Co(III) compounds which have reported Co-$N_{pyridyl}$ bond lengths of 1.91 - 1.96 Å and Co-N_{amide} bond lengths of 1.902 - 1.99 Å.[1, 3, 5, 7] These are also shorter than in related Co(II) compounds which have reported Co-$N_{pyridyl}$ bond lengths of 2.07 - 2.15 Å.[24-26] There is evidence from the crystal structure for intermolecular hydrogen bonds, these tend to be either between the amide oxygen and a neighbouring pyridyl CH group

or between a solvent molecule and the amide oxygen or pyridyl CH, with bond distances in the range 2.82 - 3.19 Å. (**Figure 2.10**). Other interactions observed in a few instances include π-π stacking interactions between aryl rings in neighbouring molecules, with π-π interactions in the range 3.64 - 3.99 Å (**Figure 2.10**).
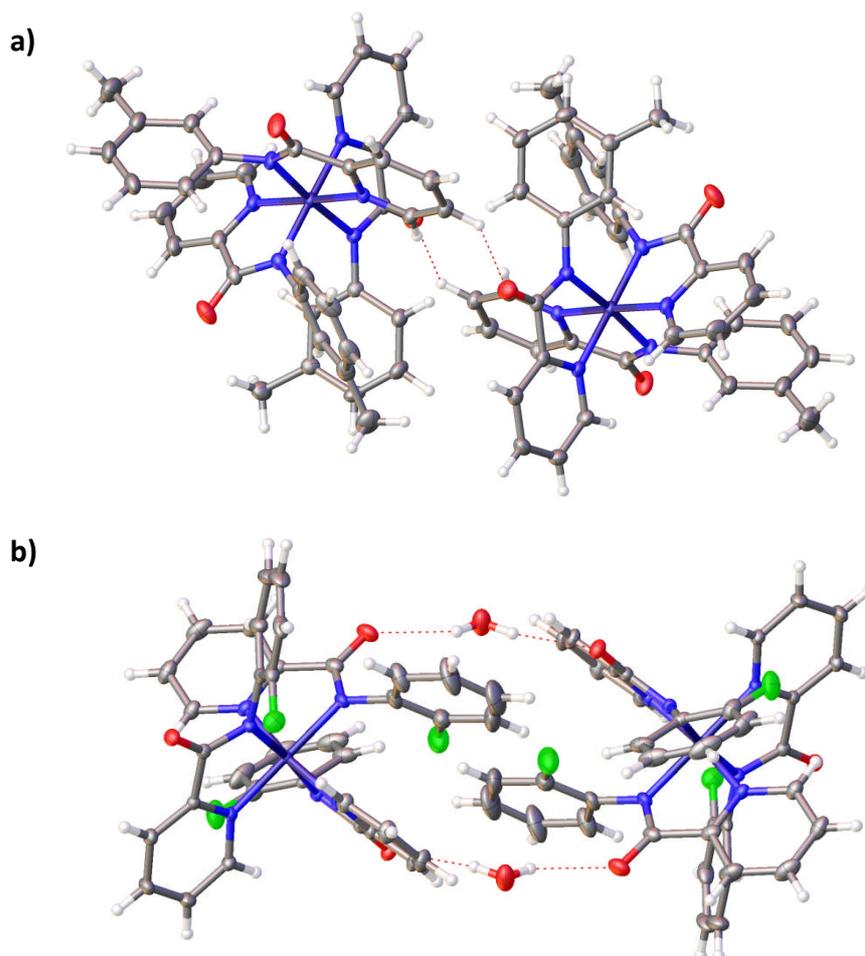
Figure 2.10: Intermolecular interactions a) hydrogen bonding between complexes b) hydrogen bonding between complex and solvent molecules and π-π stacking between complexes

2.3 Cobalt *tris*-picolinamide complexes (mixed coordination)

The cobalt picolinamide complexes synthesised within this section are shown in **Figure 2.11**. Complexes were synthesised *via* the complexation reaction of picolinamide ligand with cobalt chloride in the presence of triethylamine, similar to the synthesis of complexes with *N,N* coordination (**Chapter 2.2**) except with ethanol as the sole solvent as opposed to an ethanol-water mixture (**Scheme 2.3**).

R =   H   (**2.1b**)   3,5-diMe   (**2.16b**)
      3-Me   (**2.14b**)   4-OMe   (**2.17b**)
      4-Me   (**2.15b**)   3,5-diOMe   (**2.18b**)
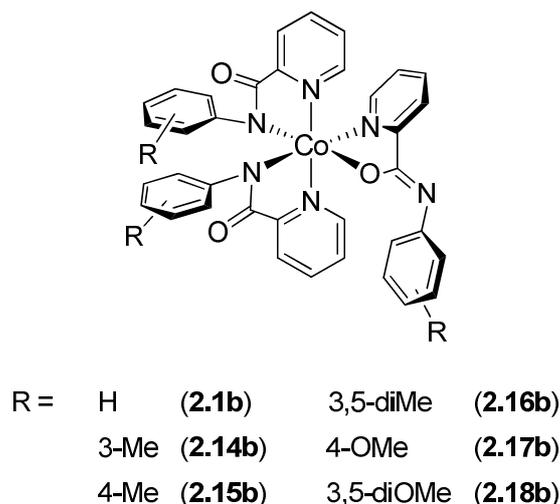
Figure 2.11: Cobalt picolinamide complexes synthesised within this section
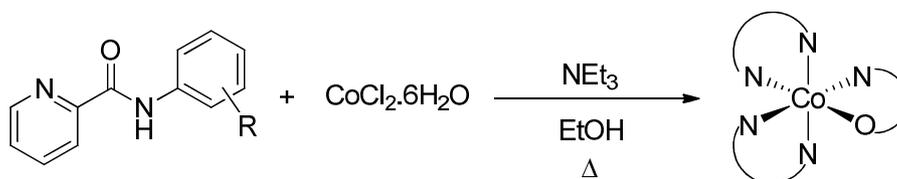
Scheme 2.3: Synthetic route for cobalt *tris*-picolinamide complexes in this section

Similar to the previous synthesis, cobalt is oxidised from Co(II) to Co(III) by triethylamine followed by the complexation of three ligands to the metal centre. Large groups or atoms e.g. chloro, methyl in the *ortho* position on the aryl ring do not result in complexation. The only difference between the synthetic pathway for these complexes compared to the previous *N,N* coordination complexes is the solvent: ethanol as opposed to a 2:1 ethanol-water mixture. This difference in solvent results in a mixture of products: the previous *N,N* coordination complex and its isomer, the mixed coordination complex shown above. These products have different retention factors which permits their separation by column chromatography to give two differently coloured complexes: red and brown

respectively. There is no interconversion between the two coordination isomers in solution over the period of one month, as evidenced by the lack of changes in the UV/vis spectra of the complexes over time (**Figure 2.12**).
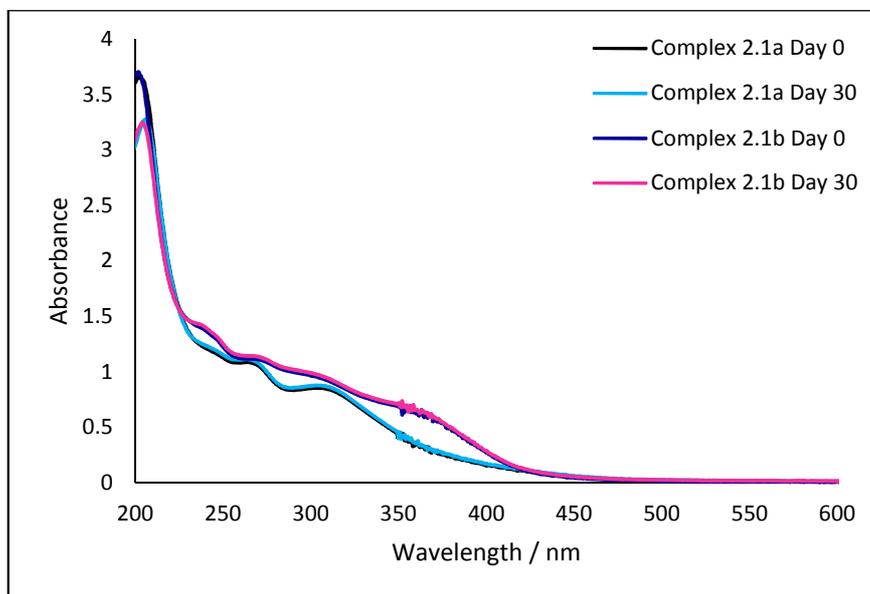
Figure 2.12: UV/vis spectra of complexes **2.1a** and **2.1b** over time

In these previously unreported mixed coordination complexes, two of the three ligands coordinate through the pyridyl nitrogen and amide nitrogen, whereas the third ligand coordinates through the pyridyl nitrogen and the amide oxygen. Combined the three ligands act as monoanionic donors to stabilise the Co(III) ion. This differs from the previously reported type of mixed coordination complexes where the *N,O* donor ligand coordination is in the neutral mode as opposed to the monoanionic mode resulting in a monocationic complex.[4] These mixed coordination complexes are only formed when electron donating groups are present as substituents on the aryl ring. When an electron donating group is present, electron density can be donated towards the amide oxygen through the conjugated π-system thus making this oxygen more nucleophilic and allowing coordination to the metal centre. However, it does not appear that this resonance effect is so great that the mixed coordination complex is the only product. In fact, even in this synthetic pathway the mixed coordination product is the minor product with approximately a 2:1 ratio in favour of the formation of the *N,N* coordination complex over the mixed coordination complex. There does not appear to be any correlation between the strength or number of electron donating substituents and the overall yield of the mixed coordination complex. Further experiments altering the solvent (toluene, acetonitrile, methanol, chloroform and

tetrahydrofuran) did not result in an increase in formation of this mixed coordination complex. Conversely, as soon as water is included in the solvent mixture the mixed coordination complex is no longer formed and only the *N,N* coordination complex is observed. The role of water in this reaction is unknown; potentially its highly polar nature allows improved stabilisation of the *N,N* coordinated ligand through hydrogen bonding and as such only the *N,N* coordination complex is formed.

2.3.1 Characterisation of cobalt *tris*-picolinamide complexes (mixed coordination)

The cobalt(III) picolinamide complexes are low spin d^6 and thus diamagnetic. The ^{1}H NMR spectra of these complexes are all very similar with the pyridyl protons (8.99 - 6.96 ppm) typically having higher chemical shifts than the aryl protons (7.50 - 5.98 ppm). Pyridyl and aryl protons tend to be shifted upfield in the complex in comparison to in the free ligand, however one set of pyridyl protons are shifted downfield. In comparison to the analogous *N,N* coordination complexes (**2.1a** - **2.18a**), a greater amount of signal overlap is observed leading to fewer peaks in the spectrum but a greater number of multiplets. Examples of the ^{1}H NMR chemical shifts for complex **2.1b** compared to the ligand are shown in **Figure 2.13** and Error! Reference source not found.. Three distinct or overlapping sets of signals for the picolinamide protons are seen in the final complex as the three ligands are inequivalent due to the slightly distorted octahedral geometry of the complex.

Table 2.5: ^{1}H NMR chemical shifts for complex **2.1b** and ligand **1.1**

Proton environment	^{1}H NMR chemical shift / ppm	
	Ligand	Complex
Pyridyl	8.62	8.99, 8.03 and 7.70
	8.31	8.43 and 7.76
	7.91	8.03 and 7.95
	8.31	7.70, 7.65 and 7.10
Aryl	7.80	6.98 and 6.92
	7.40	7.35 and 6.98
	7.16	6.98 and 6.92
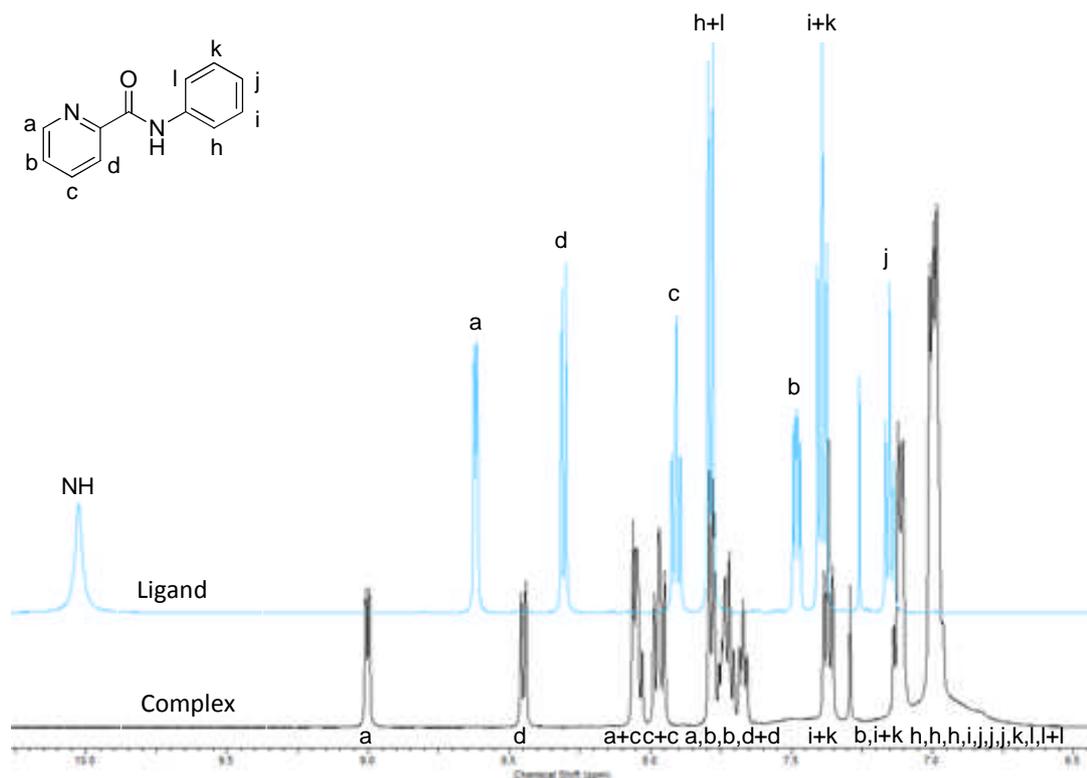
Figure 2.13: ^{1}H NMR spectra of complex **2.1b** and ligand **1.1**

The IR spectra of complexes **2.1b** - **2.18b** are similar to the equivalent *N,N* coordination isomers (**2.1a** - **2.18a**). All complexes display a weak aryl CH stretch at 3025 - 3080 cm^{-1} and a two medium intensity CO stretches at 1620 - 1640 cm^{-1} and 1560 - 1590 cm^{-1}. There are also several bands of varying intensities in the region 500 - 1600 cm^{-1}. The disappearance of the broad NH stretches (3250 - 3400 cm^{-1}) is characteristic of complex formation. The CO stretch is shifted to a lower wavenumber in the complex compared to the free ligand as is expected due to the weakening of the CO bond following coordination to the metal centre.[1-3]

The UV/vis spectrum of complex **2.1b** is shown in **Figure 2.14**. The spectrum shows an intense ligand based absorbance (π-π*) at around 200 nm with three less intense charge-transfer bounds at around 235, 270 and 300 nm. No d-d transitions are seen in the 400 - 800 nm region. The spectrum is similar to the analogous complex with *N,N* coordination (**2.1a**), the main difference being the presence of an additional charge-transfer band at around 235 nm for complex **2.1b**.[1-3]
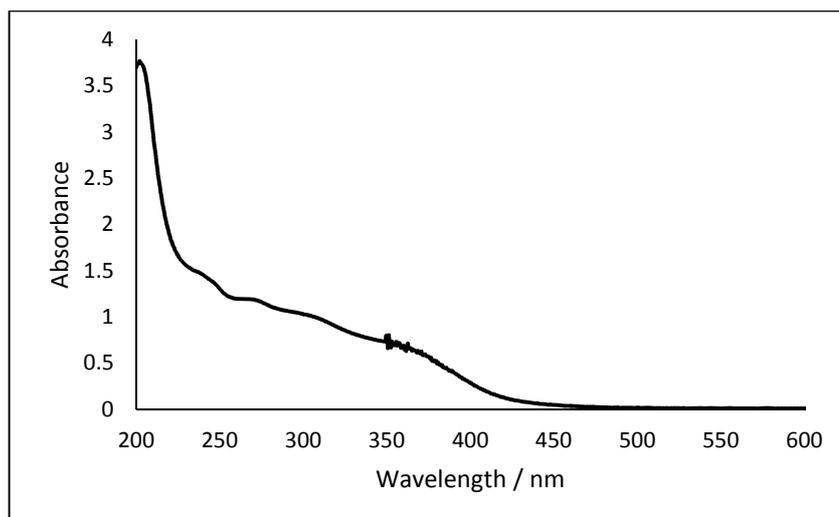
Figure 2.14: UV/vis spectrum of complex **2.1b**

2.3.2 X-Ray crystallography of cobalt *tris*-picolinamide complexes (mixed coordination)

All complexes were crystallised in a triclinic cell and structural solutions were performed in the $P\overline{1}$ space group with one (**2.1b**, **2.15b** - **2.17b**) or two (**2.14b** and **2.18b**) molecules in the asymmetric unit (**Figure 2.15**). Selected bond lengths and angles are stated in **Table 2.6**, **Table 2.7** and **Table 2.8** respectively.

2.1b   **2.14b**

2.15b   **2.16b**

2.17b   **2.18b**
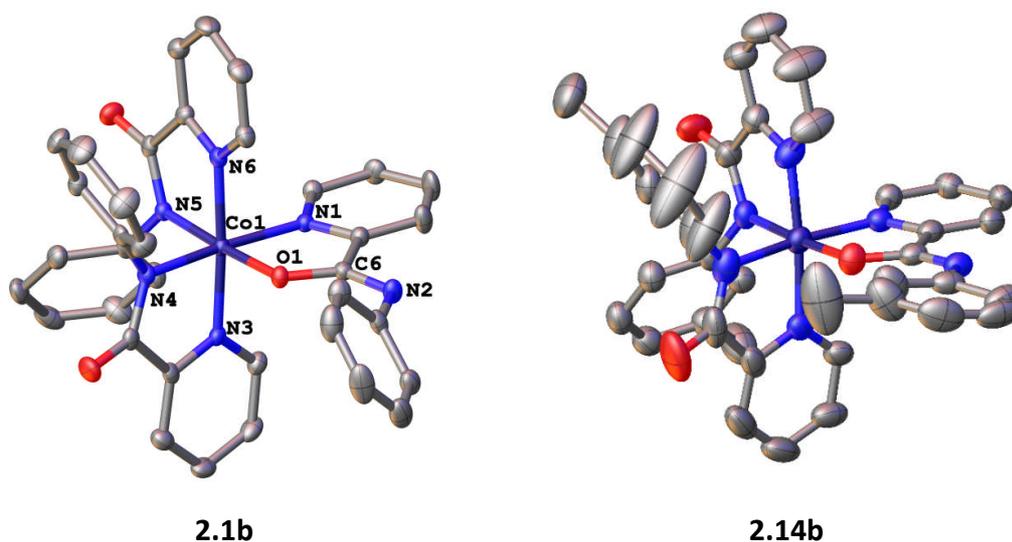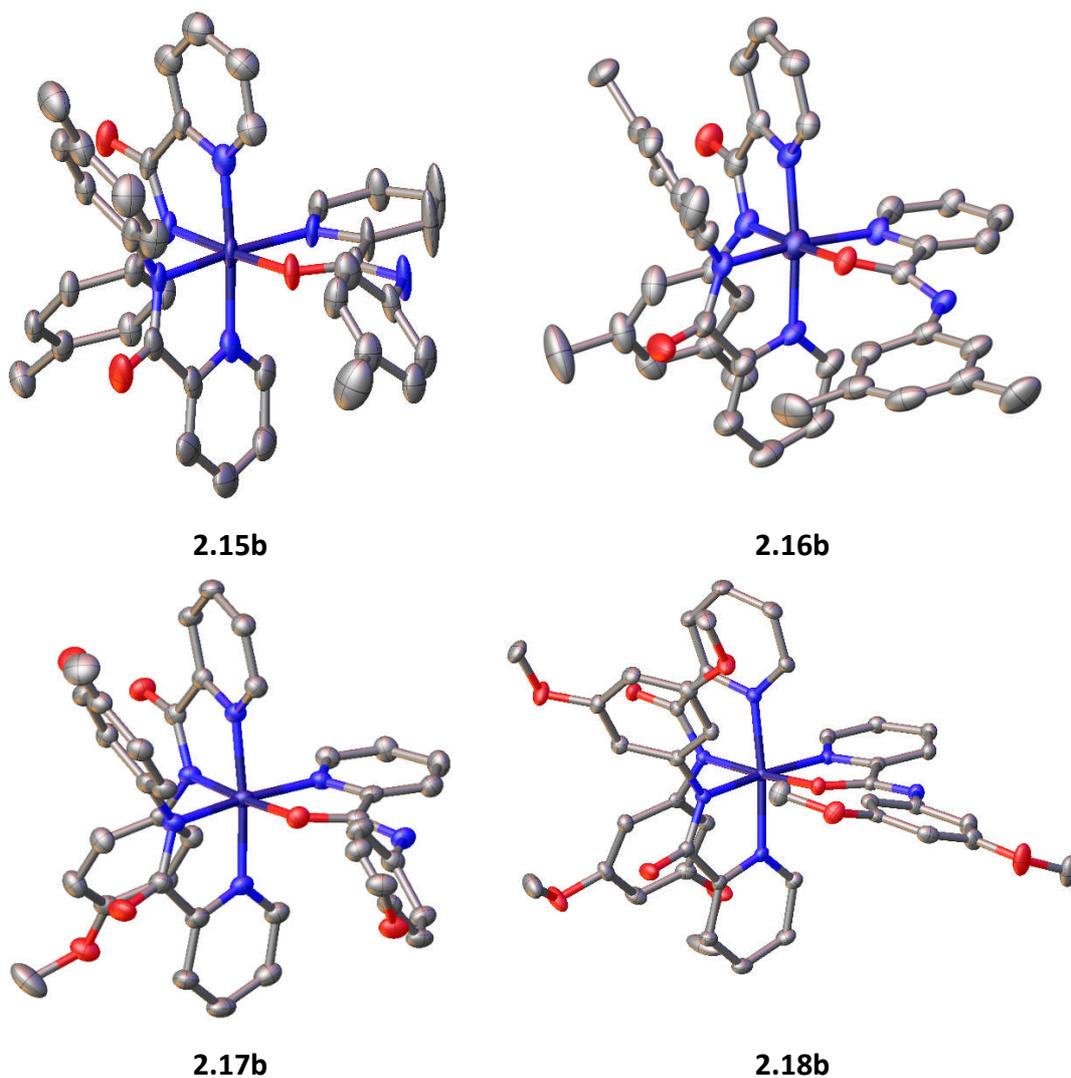
Figure 2.15: Molecular structures of complexes **2.1b** - **2.18b**. Displacement ellipsoids are at the 50 % probability level. Hydrogen atoms and co-crystallising solvent molecules are omitted for clarity. For clarity, only complex **2.1b** is labelled, the remaining complexes follow the same atom numbering scheme

Table 2.6: Selected bond lengths for complexes **2.1b** - **2.18b**

	Distance / Å					
	Co(1)-N(1)	Co(1)-N(3)	Co(1)-N(4)	Co(1)-N(5)	Co(1)-N(6)	Co(1)-O(1)
2.1b	1.959(2)	1.924(2)	1.922(2)	1.927(2)	1.921(2)	1.9154(17)
2.14b	1.927(4)	1.929(4)	1.926(4)	1.932(3)	1.949(4)	1.920(3)
2.15b	1.964(7)	1.947(9)	1.943(7)	1.929(8)	1.937(8)	1.906(6)
2.16b	1.967(4)	1.944(4)	1.929(4)	1.928(4)	1.938(4)	1.921(3)
2.17b	1.919(2)	1.933(2)	1.933(2)	1.925(2)	1.960(2)	1.9233(18)
2.18b	1.9348(19)	1.9362(19)	1.9386(19)	1.9423(19)	1.9497(19)	1.9257(16)

Table 2.7: Carbon-nitrogen bond lengths for complexes **2.1b** - **2.18b**

	Distance / Å					
	2.1b	2.14b	2.15b	2.16b	2.17b	2.18b
C(6)-N(2)	1.297(3)	1.290(6)	1.278(12)	1.284(7)	1.292(3)	1.300(3)

Table 2.8: Selected bond angles for complexes **2.1b** - **2.18b**

	Angle / °		
	N(1)-Co(1)-O(1)	O(1)-Co(1)-N(3)	O(1)-Co(1)-N(4)
2.1b	83.48(8)	88.48(8)	89.06(8)
2.14b	83.11(14)	91.05(15)	89.01(15)
2.15b	83.0(3)	89.0(3)	88.5(3)
2.16b	83.02(15)	90.78(17)	88.43(16)
2.17b	83.61(8)	90.19(9)	88.72(9)
2.18b	83.02(7)	91.53(7))	89.69(7)

As with the related *tris*-picolinamide complexes, all complexes show a distorted octahedral geometry around the central cobalt atom with the relevant bond angles in the range 82.6 - 91.3°, deviating from the expected 90° (**Table 2.8**). The cobalt-pyridyl nitrogen bond lengths are in the range 1.92 - 1.98 Å and the cobalt-amide nitrogen bond lengths are in the range 1.92 - 1.94 Å, these are

consistent with the related *tris*-picolinamide complexes with *N,N* coordination (**Chapter 2.2**). Within the *N,O* coordinated ligand, the imine carbon-nitrogen bond distance is in the range 1.26 - 1.30 Å (**Table 2.7**) and the cobalt-oxygen bond distance in the range 1.91 - 1.93 Å, these are consistent with a carbon-nitrogen double bond and a low spin cobalt(III)-oxygen single bond.[3, 27-29] There is evidence from the crystal structure for intermolecular hydrogen bonds, these tend to be between a solvent molecule and the amide oxygen or imine nitrogen, with bond distances in the range 2.93 - 3.37 Å (**Figure 2.16**). Other interactions include π-π stacking or T-stacking interactions between aryl rings in neighbouring molecules, with bond distances in the range 3.46 - 3.74 Å (**Figure 2.16**).
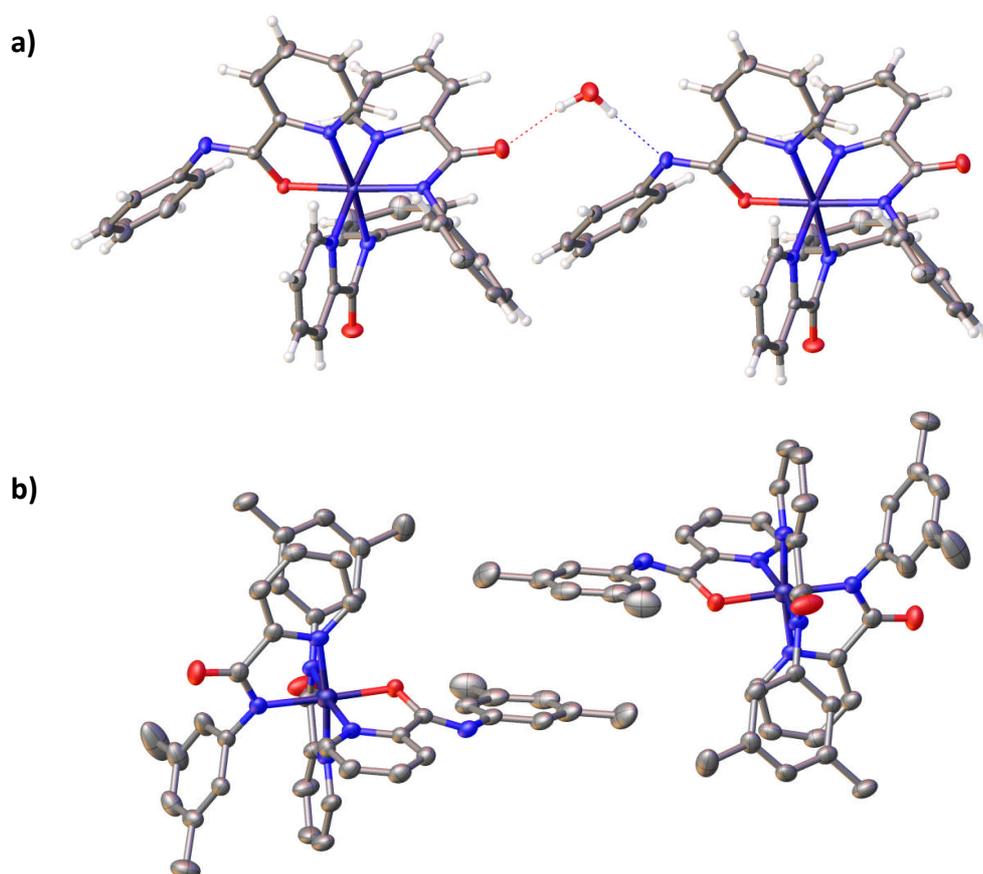
Figure 2.16: Intermolecular interactions a) hydrogen bonding between complex and solvent molecules and T-stacking between complexes b) π-π stacking between complexes

2.4 Conclusions

A range of picolinamide ligands with different electronic and steric properties have been synthesised. Utilising these ligands, a series of novel cobalt(III) *tris*-picolinamide complexes have been synthesised. The first set of complexes contain ligands which all display *N,N* coordination to the central cobalt, whereas in the second set of complexes one of the ligands instead displays *N,O* coordination. This second set of complexes is only formed with electron donating group substituents on the aryl ring and under particular solvent conditions. These two sets of complexes represent non-interconverting coordination isomers. Complexes show a distorted octahedral structure with bond lengths in keeping with a cobalt(III) species. No intramolecular bonds are observed, however intermolecular hydrogen bonds, π-π stacking and T-stacking interactions are observed between neighbouring molecules. The efficacy of the complexes as anti-cancer agents will be discussed in **Chapter 5**.

2.5 References

1.   J. Y. Qi, H. X. Ma, X. J. Li, Z. Y. Zhou, M. C. K. Choi, A. S. C. Chan and Q. Y. Yang, *Chemical Communications*, 2003, 1294-1295.

2.   A. Mishra, N. K. Kaushik, A. K. Verma and R. Gupta, *European Journal of Medicinal Chemistry*, 2008, **43**, 2189-2196.

3.   A. Ali, D. Bansal and R. Gupta, *Journal of Chemical Sciences*, 2014, **126**, 1535-1546.

4.   S. H. van Rijt, A. J. Hebden, T. Amaresekera, R. J. Deeth, G. J. Clarkson, S. Parsons, P. C. McGowan and P. J. Sadler, *Journal of Medicinal Chemistry*, 2009, **52**, 7753-7764.

5.   M. Ray, D. Ghosh, Z. Shirin and R. Mukherjee, *Inorganic Chemistry*, 1997, **36**, 3568-3572.

6.   N. K. Kaushik, A. Mishra, A. Ali, J. S. Adhikari, A. K. Verma and R. Gupta, *Journal of Biological Inorganic Chemistry*, 2012, **17**, 1217-1230.

7.   A. Mishra, A. Ali, S. Upreti and R. Gupta, *Inorganic Chemistry*, 2008, **47**, 154-161.

8.   A. P. Singh, A. Ali and R. Gupta, *Dalton Transactions*, 2010, **39**, 8135-8138.

9.   A. P. Singh, G. Kumar and R. Gupta, *Dalton Transactions*, 2011, **40**, 12454-12461.

10.   L. A. Tyler, M. A. Olmstead and P. K. Mascharak, *Inorganic Chemistry*, 2001, **40**, 5408-5414.

11.   A. J. Canty and C. V. Lee, *Inorganica Chimica Acta-Letters*, 1981, **54**, L205-L206.

12.   T. J. Collins, *Accounts of Chemical Research*, 1994, **27**, 279-285.

13.   D. O'Hagan, *Journal of Fluorine Chemistry*, 2010, **131**, 1071-1081.

14.   J. Wang, M. Sanchez-Rosello, J. L. Acena, C. del Pozo, A. E. Sorochinsky, S. Fustero, V. A. Soloshonok and H. Liu, *Chemical Reviews*, 2014, **114**, 2432-2506.

15.   T. Cheng, W. Y. Yin, Y. Zhang, Y. N. Zhang and Y. Huang, *Org. Biomol. Chem.*, 2014, **12**, 1405-1411.

16.   M. Colombo, S. Bossolo and A. Aramini, *Journal of Combinatorial Chemistry*, 2009, **11**, 335-337.

17.   D. W. Engers, C. M. Niswender, C. D. Weaver, S. Jadhav, U. N. Menon, R. Zamorano, P. J. Conn, C. W. Lindsley and C. R. Hopkins, *Journal of Medicinal Chemistry*, 2009, **52**, 4115-4118.

18.   S. K. Kang, D. W. Kim and J. N. Park, *Synlett*, 2002, 427-430.

19.   A. M. Martinez, N. Rodriguez, R. G. Arrayas and J. C. Carretero, *Chemical Communications*, 2014, **50**, 2801-2803.

20.   C. Sambiagio, R. H. Munday, S. P. Marsden, A. J. Blacker and P. C. McGowan, *Chem.-Eur. J.*, 2014, **20**, 17606-17615.

21.   S. Dutta, S. Pal and P. K. Bhattacharya, *Polyhedron*, 1999, **18**, 2157-2162.

22.   L. Huang, Q. Li, C. Wang and C. Qi, *Journal of Organic Chemistry*, 2013, **78**, 3030-3038.

23.   C. E. Housecroft and A. G. Sharpe, *Inorganic Chemistry*, Fourth edn., Pearson Ltd., 2012.

24.   M. Dakovic, D. Vila-Vicosa, M. J. Calhorda and Z. Popovic, *CrystEngComm*, 2011, **13**, 5863-5871.

25.   G. V. Tsintsadze, R. A. Kiguradze, A. N. Shnulin and K. S. Mamedov, *Journal of Structural Chemistry*, 1986, **27**, 272-278.

26.   J. Xue, X. Hua, L. Yang, W. Li, Y. Xu, G. Zhao, G. Zhang, L. Liu, K. Liu, J. e. Chen and J. Wu, *Journal of Molecular Structure*, 2014, **1059**, 108-117.

27.   F. H. Allen, O. Kennard, D. G. Watson, L. Brammer, A. G. Orpen and R. Taylor, *Journal of the Chemical Society-Perkin Transactions 2*, 1987, S1-S19.

28.   A. Böttcher, T. Takeuchi, K. I. Hardcastle, T. J. Meade, H. B. Gray, D. Cwikel, M. Kapon and Z. Dori, *Inorganic Chemistry*, 1997, **36**, 2498-2504.

29.   K. Neuvonen, F. Fülöp, H. Neuvonen, A. Koch, E. Kleinpeter and K. Pihlaja, *Journal of Organic Chemistry*, 2003, **68**, 2151-2160.

Chapter 3

Synthesis and characterisation of cobalt (II) *bis*-picolinamide complexes

3. Cobalt (II) *bis*-picolinamide complexes

Cobalt(II) *bis*-picolinamide complexes have been sparingly studied so far, with only a handful of structural studies published in the literature. The first study was carried out by Mamedov, investigating how the position of the amide group from the pyridine ring influences the structure of the corresponding metal complexes.[1] Structural data showed dicationic octahedral cobalt(II) complexes with two picolinamide ligands which coordinate in the neutral *N,O* mode, with two aqua ligands arranged in the *trans* geometry and two acetate counterions (**Figure 3.1a**). Further studies with chloride or squarate acting as the counterion confirmed this *trans* geometry.[2, 3] Corresponding nickel(II) and zinc(II) aqua complexes also display the *trans* geometry.[4] Alteration of the axial aqua ligand to thiocyanate produced a neutral cobalt complex and altered the geometry with the thiocyanate ligands arranged in the *cis* geometry, although the pyridyl rings remain *trans* to each other (**Figure 3.1b**).[5] Density functional theory (DFT) calculations found that the lowest energy form of this complex involved the picolinamide ligands in the *cis* geometry and the thiocyanate ligands coordinating through the nitrogen, as opposed to the sulfur atom. Corresponding nickel(II) and zinc(II) thiocyanate complexes also display the *cis* geometry. However, the analogous copper(II) complex contains the two thiocyanate ligands in the *trans* geometry and coordinated to the central copper through the sulfur atom. The biological activity of such cobalt(II), copper(II), nickel(II) and zinc(II) *bis*-picolinamide complexes has not been investigated. However, the corresponding zinc(II) complex with axial aqua ligands and nitrate counterions showed a small tendency to decrease monocyte and neutrophil intracellular killing capacity by approximately 5 %.[4]
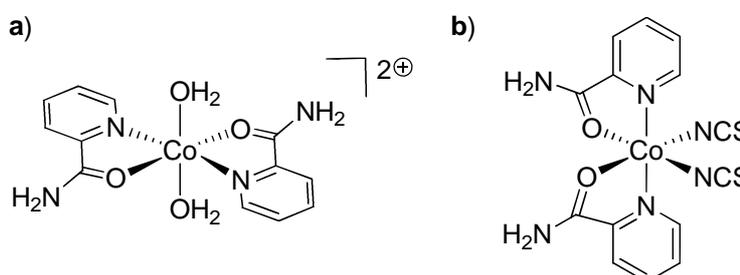
Figure 3.1: Previously published cobalt(II) picolinamide complexes displaying varying *cis* and *trans* geometries

3.1 Cobalt *bis*-picolinamide thiocyanate complexes

The cobalt picolinamide complexes synthesised within this section are shown in **Figure 3.2**. Complexes were synthesised *via* the complexation reaction of picolinamide ligand with cobalt nitrate in the presence of potassium thiocyanate, based on the synthesis reported by Đaković *et al.* (**Scheme 3.1**).[5]

R = H   (**3.1**)   2-Me   (**3.4**)
     3-Me   (**3.2**)   4-Me   (**3.5**)
     3,5-diMe   (**3.3**)   2,3-diMe (**3.6**)
                                         4-OMe   (**3.7**)
                                         3,5-diOMe (**3.8**)

Figure 3.2: Cobalt picolinamide complexes synthesised within this section

Scheme 3.1: Synthetic route for cobalt *bis*-picolinamide complexes in this section

In this synthesis, the two ligands coordinate through the pyridyl nitrogen and amide oxygen, acting as neutral donors to help stabilise the 2+ oxidation state of the central cobalt ion, as has been reported previously.[1, 2, 5] The axial thiocyanate ligands coordinate to the cobalt through the nitrogen atom, as opposed to through the sulfur atom; this result is consistent with the previous complexes and DFT

calculations performed by Đaković *et al.*[5] The synthesis was only successful when there were no electron withdrawing groups as substituents on the aryl ring. This may be explained by the increased acidity of the amide proton when electron withdrawing groups are present which may hinder *N,O* coordination to the metal centre. This is consistent with previous results for cobalt(III) *tris*-picolinamide complexes where the nature of the aryl ring substituents impacted on the coordination modes of the picolinamide ligand (**Chapter 2**).

Each cobalt *bis*-picolinamide complex may form five potential geometric isomers (Error! Reference source not found.). However, only the *trans-trans-trans* and *cis-trans-cis* isomers were observed. In general, when the aryl ring is unsubstituted or substituted in the *meta* position, the thiocyanate molecules are positioned *trans* to each other; whereas substitution in the *ortho* or *para* positions on the aryl ring result in the corresponding *cis* complexes. However, complex **3.8**, containing the 3,5-dimethyoxy picolinamide ligand, does not obey this general rule and is observed in the *cis* geometry rather than the expected *trans* geometry. The geometry of complex **3.6** is unknown, however it is hypothesised to adopt the *cis* geometry due to the presence of an *ortho* substituent on the picolinamide aryl ring.

If substitution in the *ortho* position has a greater effect than substitution in the *meta* position this may explain why attempts to synthesise the cobalt(II) *bis*-picolinamide complex with the anthracene picolinamide ligand (**1.22**) were unsuccessful. The bulky nature of this ligand presumably would prevent the formation of the *cis* geometry due to steric hindrance. The majority of cobalt(II) *bis*-picoliamide complexes previously reported contain axial ligands in the *trans* position. Differences in *cis*/*trans* geometry have been noted with alterations in the axial ligand (aqua[1] to thiocyanate[5]) and the metal ion (cobalt to copper[5]), though the nature of the axial ligand appears to be the more important factor. All previous studies have involved the picolinamide ligand with no N-aryl substitutions and thus this represents the first report on the geometric effects of changing the picolinamide ligand substituents. This work implies that the effect of ligand substituents on geometry is greater than the effect of the type of axial ligand. It is suggested that differing geometries have differing thermodynamic stabilities depending upon the N-aryl substitution, although the reason for these differences is unclear. The fact that altering the position of the same group, e.g. methyl, alters the geometry perhaps implies that steric effects are of greater importance than electronic effects. Perhaps α, β, γ C-H interactions between the aryl ring and the axial ligands are important during the transition phase to help stabilise one

geometry over the other.[6] Alternatively, intra-ligand n to π* transitions or hydrogen bonding may also assist the stabilisation of one particular geometry.[7, 8]
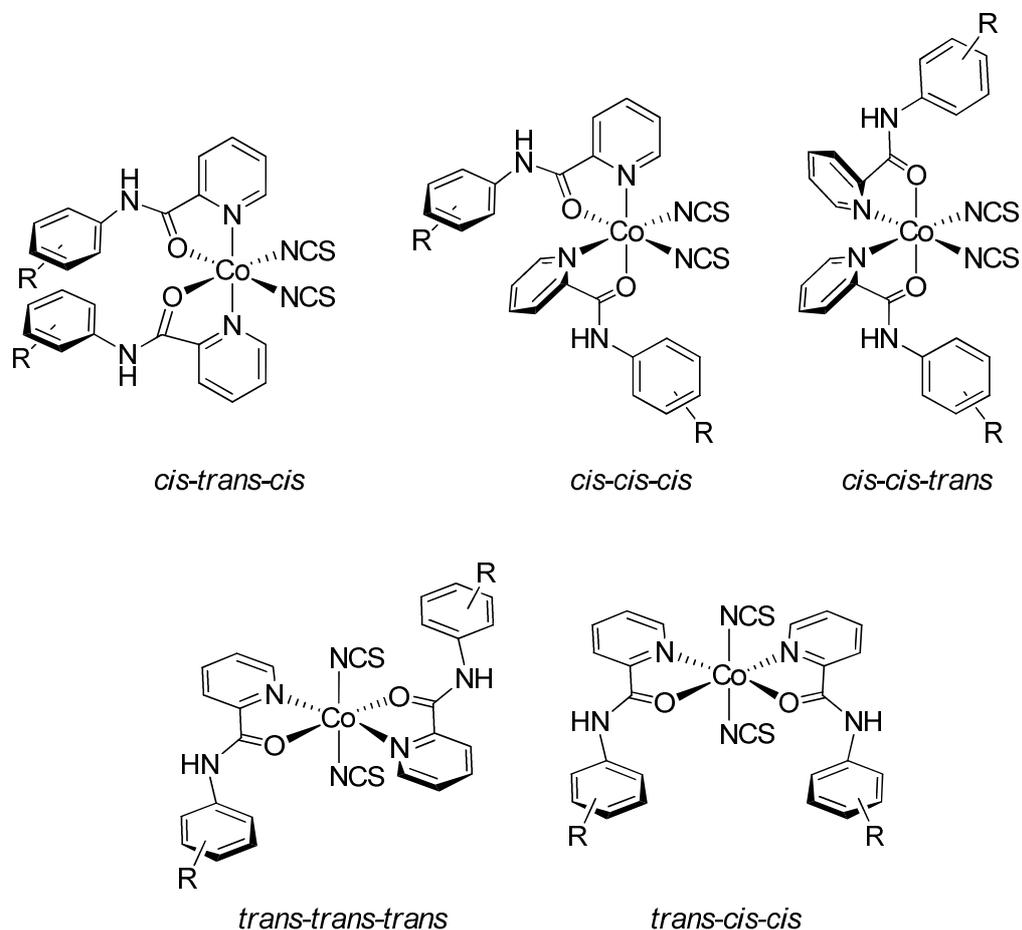
Figure 3.3: Potential isomers of cobalt *bis*-picolinamide thiocyanate complexes. Geometric (*cis / trans*) descriptors are designated in the order: thiocyanate ligand, picolinamide pyridyl ring, picolinamide amide group

3.1.1 Characterisation of cobalt *bis*-picolinamide thiocyanate complexes

The cobalt *bis*-picolinamide complexes contain a central cobalt(II) ion which is paramagnetic (d[7]). Therefore the ^{1}H NMR spectra of complexes **3.1** - **3.8** show a number a broad peaks; however the pattern of peaks is the same for the complexes as for the free ligands and thus assignment is possible. The ^{1}H NMR spectra of these complexes are all very similar with the amide proton in the region 10.54 - 10.20 ppm and the pyridyl protons (8.70 - 7.45 ppm) typically having slightly higher chemical shifts than the aryl protons (7.96 - 6.21 ppm). All pyridyl and aryl protons are shifted slightly upfield in the complex compared to in the free ligand. Examples of the ^{1}H NMR chemical shifts for complex **3.4** compared to the ligand are shown in **Figure 3.4** and **Table 3.1**.
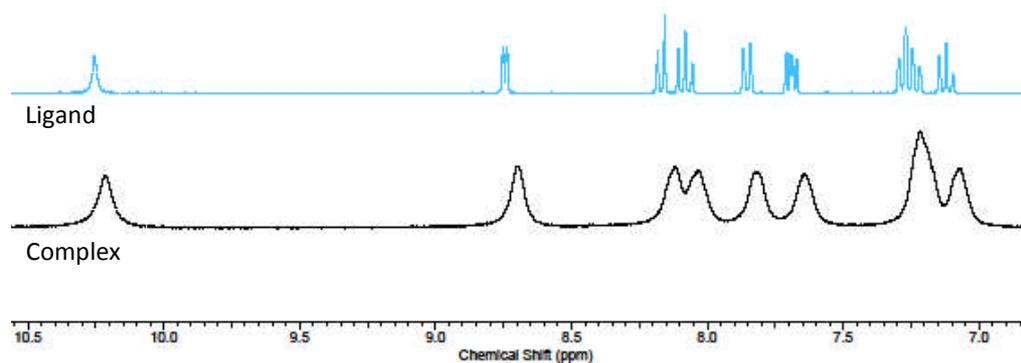
Figure 3.4: ^{1}H NMR of complex **3.4** and ligand **1.15**

Table 3.1: ^{1}H NMR chemical shifts for complex **3.4** and ligand **1.15**

Proton environment	^{1}H NMR chemical shift / ppm	
	Ligand	**Complex**
Amide	10.26	10.21
Pyridyl	8.75	8.70
	8.17	8.12
	8.08	8.03
	7.70	7.64
Aryl	7.86	7.82
	7.27	7.22
	7.12	7.07

The IR spectra of complexes **3.1** - **3.8** all display a broad NH stretch at 3200 - 3300 cm^{-1}, a weak aryl CH stretch at 3050 - 3100 cm^{-1}, a strong CN (from NCS) at 2070 - 2100 cm^{-1} and a strong CO stretch at 1610 - 1635 cm^{-1}. There are also several bands of varying intensities in the region 500 - 1600 cm^{-1}. The CO stretch is shifted to a lower wavenumber in the complex compared to the free ligand as is expected due to the weakening of the CO bond following coordination to the metal centre. The CN stretch from the thiocyanate ligand is diagnostic for the formation of the cobalt *bis*-picolinamide thiocyanate complex.[2, 3, 5]

The UV/vis spectrum of complex **3.1** is shown in **Figure 3.5**. The spectrum shows an intense ligand based absorbance (π-π*) at around 200 nm with two less intense

charge-transfer bounds at around 220 and 280 nm and finally a weak d-d transition at around 640 nm.[2, 3]
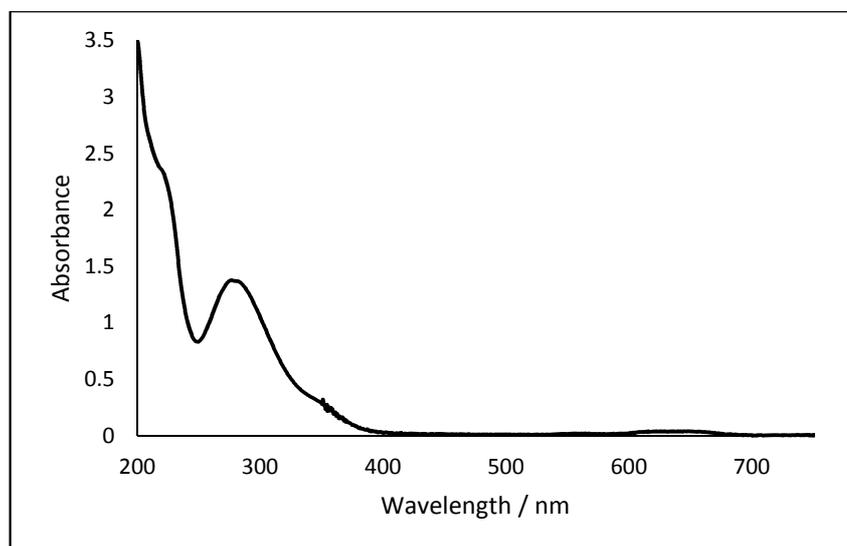
Figure 3.5: UV/vis spectrum of complex **3.1**

3.1.2 X-Ray crystallography of cobalt *bis*-picolinamide thiocyanate complexes

Complexes in the *trans* geometry (**3.1** - **3.3**) were crystallised in a monoclinic (**3.1**, **3.2**) or triclinic cell (**3.3**) and structural solutions were performed in the $P\overline{1}$ (**3.1**, **3.3**) or $P\ 2_1/c$ (**3.2**) space group with half (**3.1, 3.2, 3.3**) a molecule in the asymmetric unit (**Figure 3.6**). Selected bond lengths and angles are stated in **Table 3.2** and **Table 3.3** respectively.

3.1   **3.2**

3.3
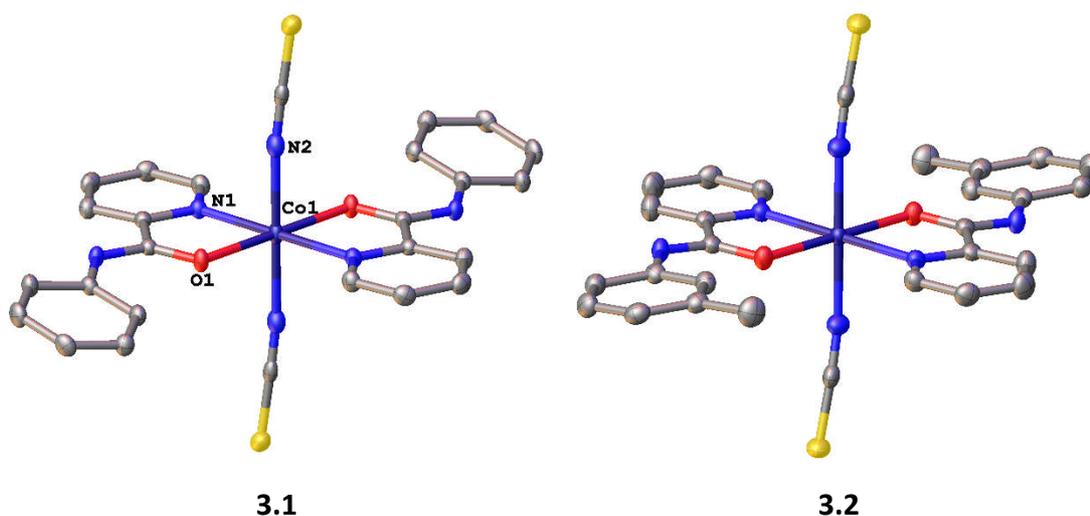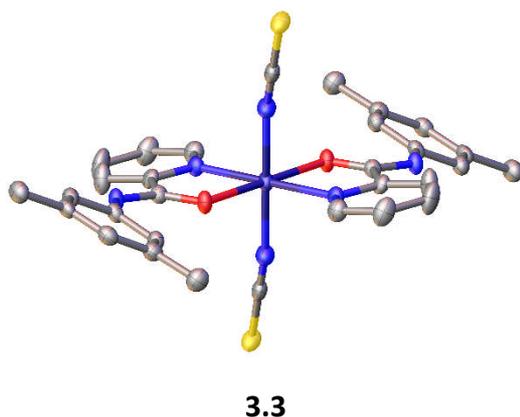
Figure 3.6: Molecular structures of complexes **3.1** - **3.3**. Displacement ellipsoids are at the 50 % probability level. Hydrogen atoms are omitted for clarity. For clarity, only complex **3.1** is labelled, the remaining complexes follow the same atom numbering scheme

Table 3.2: Selected bond lengths for complexes **3.1** - **3.3**. [a] = symmetry generated

Distance / Å	Complex		
	3.1	**3.2**	**3.3**
Co(1)-N(1)	2.1064(15)	2.117(2)	2.114(3)
Co(1)-N(1)'	2.1064(15)[a]	2.117(2)[a]	2.114(3)[a]
Co(1)-O(1)	2.1021(12)	2.0934(17)	2.124(2)
Co(1)-O(1)'	2.1021(12)[a]	2.0934(17)[a]	2.124(2)[a]
Co(1)-N(2)	2.0827(17)	2.070(2)	2.060(3)
Co(1)-N(2)'	2.0827(17)[a]	2.070(2)[a]	2.060(3)[a]

Table 3.3: Selected bond angles for complexes **3.1** - **3.3**

	Angle / °		
	N(1)-Co(1)-O1	**N(1)-Co(1)-N(2)**	**O(1)-Co(1)-N(1)'**
3.1	77.28(5)	89.32(6)	102.72(5)
3.2	76.86(7)	91.32(9)	103.14(7)
3.3	76.81(9)	86.37(10)	103.19(9)

All complexes show a distorted octahedral geometry around the central cobalt atom with the relevant bond angles in the range 76.8 - 91.3°, deviating from the expected 90° (**Table 3.3**). This distortion can be ascribed to the restrictions imposed by the bidentate picolinamide ligand. The cobalt-pyridyl nitrogen bond lengths are in the range 2.10 - 2.12 Å, the cobalt-oxygen bond lengths are in the range 2.09 - 2.12 Å and the cobalt-thiocyanate nitrogen bond lengths are in the range 2.06 - 2.08 Å. This is consistent with related high spin Co(II) compounds which have reported Co-$N_{pyridyl}$ bond lengths of 2.07 - 2.12 Å and Co-O_{amide} bond lengths of 2.06 - 2.12 Å and the one related Co(II) thiocyanate compound which has a reported Co-$N_{thiocyanate}$ bond length of 2.07 Å.[1-3, 5] These are also longer than in related Co(III) compounds which have reported Co-$N_{pyridyl}$ bond lengths of 1.91 - 1.96 Å.[9-12] There is evidence from the crystal structure for an intramolecular hydrogen bond between the amide oxygen and the aryl *ortho*-CH group of the same ligand, with the bond distance in the range 2.83 - 2.86 Å. There is also evidence for an intermolecular hydrogen bond between the thiocyanate sulfur and the neighbouring amide NH group, with the bond distance in the range 3.55 - 3.61 Å. Other interactions observed include π-π stacking interactions between aryl rings and pyridyl rings in neighbouring molecules, with π-π interaction distances in the range 3.65 - 3.87 Å, and in the case of complex **3.1** alignment of the thiocyanate groups (**Figure 3.7**).
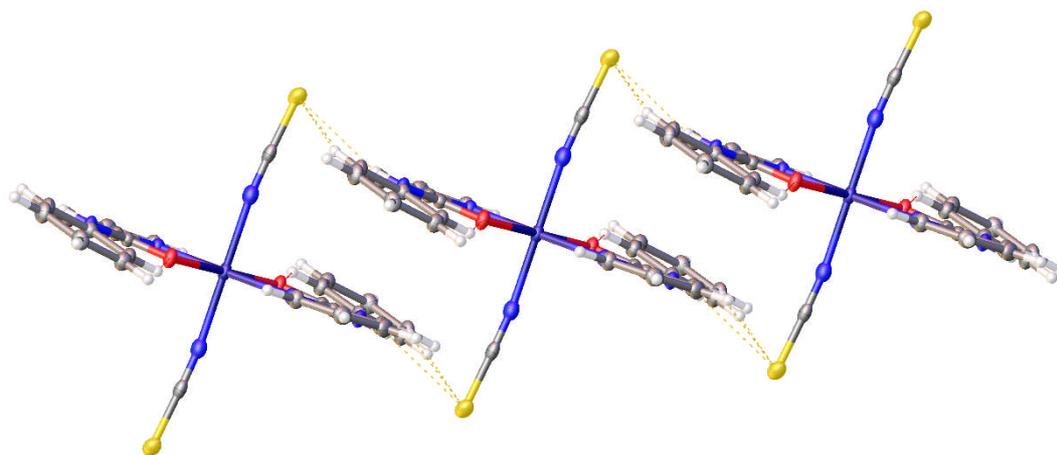
Figure 3.7: Intermolecular hydrogen bonding and π-π stacking interactions

Complexes in the *cis* geometry (**3.4** - **3.8**) were crystallised in a monoclinic (**3.4**, **3.5, 3.7**) or triclinic (**3.8**) cell and structural solutions were performed in the *P* 2₁/c (**3.4**), *Cc* (**3.5, 3.7**) or *P* $\overline{1}$ (**3.8**) space group with one (**3.4**, **3.5, 3.7**) or four (**3.8**) molecules in the asymmetric unit (**Figure 3.8**). Selected bond lengths and angles are stated in **Table 3.4** and respectively **Table 3.5**.

3.4   **3.5**

3.7   **3.8**
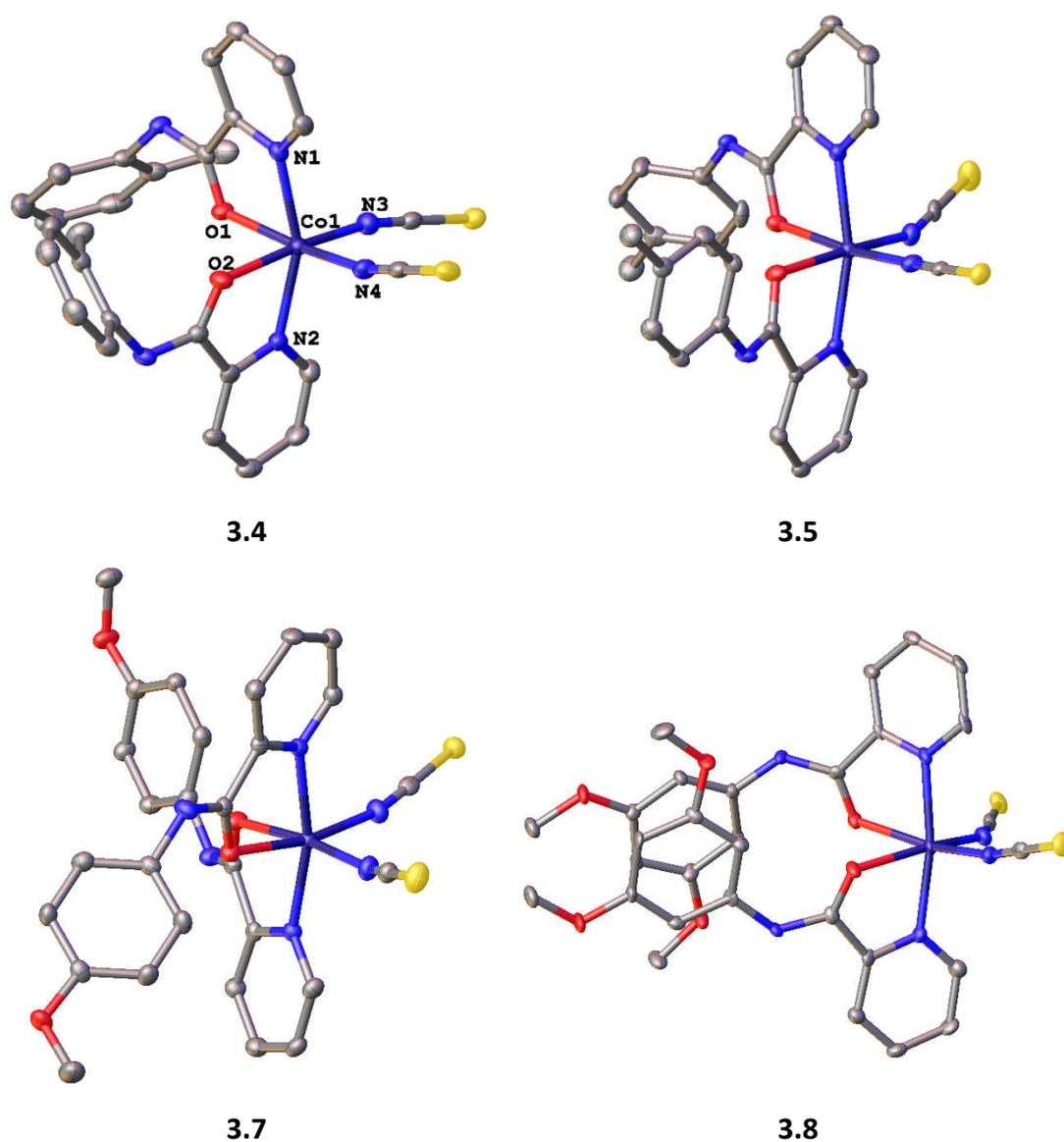
Figure 3.8: Molecular structures of complexes **3.4** - **3.8**. Displacement ellipsoids are at the 50 % probability level. Hydrogen atoms are omitted for clarity. For clarity, only complex **3.4** is labelled, the remaining complexes follow the same atom numbering scheme

Table 3.4: Selected bond lengths for complexes **3.4** - **3.8**

Distance / Å	Complex			
	3.4	**3.5**	**3.7**	**3.8**
Co(1)-N(1)	2.135(3)	2.127(3)	2.129(2)	2.112(4)
Co(1)-N(2)	2.141(3)	2.110(3)	2.102(2)	2.127(4)
Co(1)-O(1)	2.122(3)	2.133(2)	2.144(2)	2.231(4)
Co(1)-O(2)	2.138(3)	2.155(2)	2.165(2)	2.194(4)
Co(1)-N(3)	2.069(4)	2.071(3)	2.080(3)	2.061(5)
Co(1)-N(4)	2.048(4)	2.046(3)	2.048(3)	2.054(5)

Table 3.5: Selected bond angles for complexes **3.4** - **3.8**

	Angle / °		
	N(1)-Co(1)-O(1)	**O(1)-Co(1)-N(3)**	**N(3)-Co(1)-N(4)**
3.4	75.90(12)	89.42(14)	94.87(15)
3.5	75.94(11)	87.46(11)	99.13(13)
3.7	76.10(9)	87.38(10)	99.49(11)
3.8	75.32(15)	89.47(18)	101.7(2)

All complexes show a distorted octahedral geometry around the central cobalt atom with the relevant bond angles in the range 75.3 - 99.5°, deviating from the expected 90° (**Table 3.5**). This distortion can be ascribed to the restrictions imposed by the bidentate picolinamide ligand. The *cis* geometry complexes show greater distortion from an octahedral compared to the *trans* geometry complexes, evidenced by the distortion of the bond angle between the two thiocyanate nitrogen atoms from the expected 90° by up to 9.5°. This distortion is similar to the previous cobalt *bis*-picolinamide thiocyanate complex in the *cis* geometry, where the bond angle between the two thiocyanate nitrogen atoms was reported as 96.9°.[5] The cobalt-pyridyl nitrogen bond lengths are in the range 2.10 - 2.14 Å, the cobalt-oxygen bond lengths are in the range 2.12 - 2.23 Å and the cobalt-thiocyanate nitrogen bond lengths are in the range 2.04 - 2.08 Å. There is evidence from the crystal structure for an intermolecular hydrogen bond between the thiocyanate sulfur and the neighbouring amide NH group, with bond distances in the range 3.32 - 3.50 Å. Other interactions observed include π-π stacking

interactions between aryl rings and pyridyl rings in neighbouring molecules, with π-π interaction distances in the range 3.55 - 3.83 Å (**Figure 3.9**).
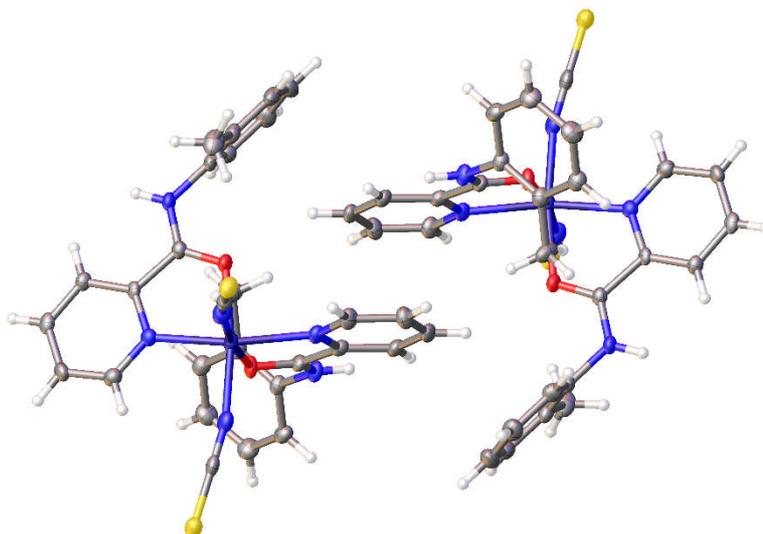
Figure 3.9: Intermolecular π-π stacking interactions

3.1.3 Powder X-ray diffraction of cobalt *bis*-picolinamide thiocyanate complexes

Powder X-ray diffraction patterns of complexes **3.1** - **3.8** were recorded to allow comparison with the single crystal data. In the solid state, complexes **3.1** - **3.3** adopt the *trans* geometry whereas complexes **3.4** - **3.8** adopt the *cis* geometry. Experimental powder diffraction data can be compared to powder diffraction data simulated from the single crystal structure to determine whether the bulk sample is consistent with the single crystals i.e. whether a single isomer or a mixture of isomers is present. Powder diffraction data for complexes **3.1** - **3.8** is displayed in **Figure 3.10**. Due to the lack of a suitable crystal structure of complex **3.6**, no simulated powder diffraction data is presented.

Figure 3.10: X-ray powder diffraction patterns for complexes **3.1** - **3.3** (*trans*) and **3.4** - **3.8** (*cis*)

Key: Black = simulated, blue = experimental

All the *trans* complexes (**3.1** - **3.3**) show good agreement between the experimental and the simulated powder diffraction patterns, with all the high intensity peaks from the simulated pattern present at similar 2θ positions in the experimental patterns. This confirms that the bulk samples of complexes **3.1** - **3.3** consist of only the *trans* isomer. For the *cis* complexes with simulated data (**3.4**, **3.5**, **3.7**, **3.8**) there are some similarities but also some differences between the experimental and the simulated powder diffraction patterns. The high intensity peaks from the simulated pattern do appear in the experimental patterns, but some other peaks are also present in the experimental diffraction pattern which cannot be matched to the corresponding simulated pattern. This could be ascribed to the presence of a mixture of isomers in the bulk powder sample compared to the single crystals analysed or could be due to differences in the solvent content between the crystal and powder samples. If the observed differences are due to the presence of a mixture of isomers, as this is only observed for *cis* complexes, this may imply that the *trans* complex is the thermodynamic product and the *cis* complex (or mixture) is the kinetic product of the complex synthesis.

3.1.4 Magnetic studies of cobalt *bis*-picolinamide thiocyanate complexes

Cobalt(II) has d^7 electron configuration and can exist in either a low spin or a high spin state (**Figure 3.11**). The calculated spin only magnetic moment for low spin Co(II) is 1.73 μ_B with the observed values generally in the range 1.8 - 2.2 μ_B; the calculated spin only magnetic moment for a high spin Co(II) is 3.88 μ_B with the observed values generally in the range 4.3 - 5.2 μ_B.[13] The chemical environment around the central cobalt ion affects which spin state is adopted. Octahedral cobalt(II) complexes tend to be high spin but transitions between the two spin states are known to occur, this phenomenon is known as spin crossover (SCO). SCO in cobalt(II) complexes is uncommon and often gradual or incomplete; however there are examples of ligand systems which display abrupt SCO with thermal hysteresis, these ligands tend to be based upon the terpyridine ligand system.[14-19]

low spin
^{2}Co(II) S=$^1/_2$

high spin
^{4}Co(II) S=$^3/_2$

Figure 3.11: Low and high spin states of cobalt(II)

Cobalt(II) *bis*-picolinamide complexes have previously been shown to be high spin (μ_{eff} = 4.1 μ_B).[3] The magnetic behaviour of complex **3.4** was investigated in both the solution and solid states. Complex **3.4** was selected as it displays the highest solubility. In the solution state, the magnetic moment is measured *via* the Evans NMR method.[20] In the solid state, the magnetic moment as a function of temperature is measured by a superconducting quantum interference device (SQUID).[21] In the solution state, the magnetic moment for complex **3.4** was found to be 4.01 μ_B at 300 K in d_3-acetonitrile. Due to limited solubility the Evans NMR method was only performed at a single temperature. This is consistent with a high spin central cobalt(II) ion in solution. The magnetic moment is lower than may be expected due to the low solubility of the complex; if the concentration of the solution was altered before the measurement performed (due to some of the complex precipitating out of solution) this would affect the magnetic moment calculation. The solid state magnetic moment measurements for complex **3.4**, collected by Dr. Rafal Kulmaczewski at The University of Leeds using a SQUID magnetometer in a field of 5 kG, are shown in **Figure 3.12**. The magnetic moment is shown to be 4.36 μ_B from 300 - 75 K before dropping at lower temperatures due to the effect of zero field splitting. This supports the solution state data and again shows a high spin cobalt(II) ion which does not undergo spin transitions.
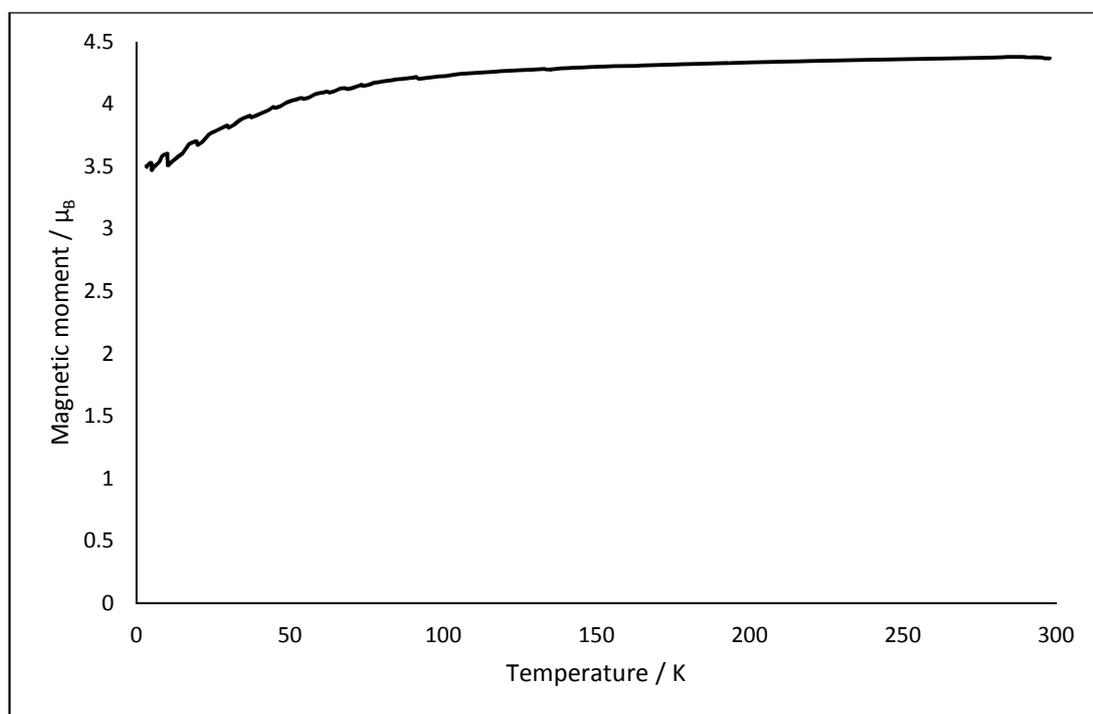
Figure 3.12: Variable temperature magnetic moment measurements of complex **3.4**

3.2 Adaptations of cobalt *bis*-picolinamide complexes

The cobalt picolinamide complexes synthesised within this section are shown in **Figure 3.13**. Complexes were synthesised *via* the complexation reaction of picolinamide ligand either with cobalt chloride or with cobalt nitrate in the presence of potassium iodide, adapted from the synthesis reported by Đaković *et al*. (**Scheme 3.2**).[5]

(3.9)   (3.10)
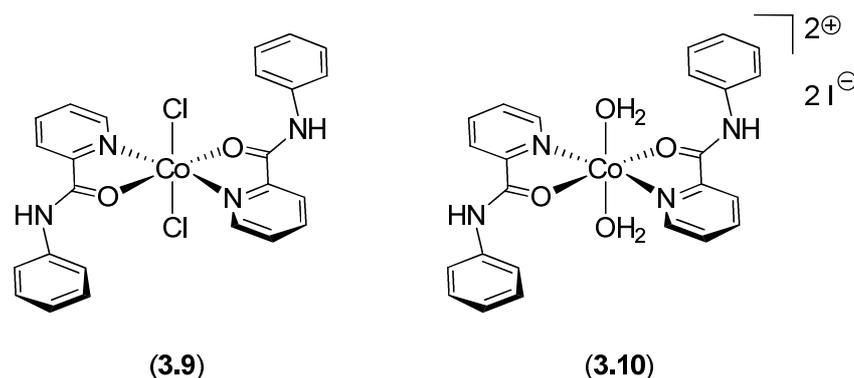
Figure 3.13: Cobalt picolinamide complexes synthesised within this section
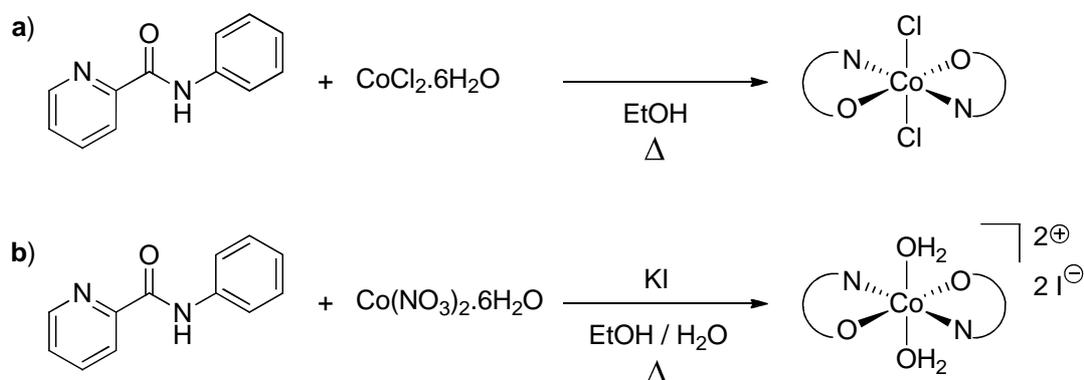
Scheme 3.2: Synthetic route for cobalt *bis*-picolinamide complexes in this section

Similar to the cobalt *bis*-picolinamide thiocyanate complexes (**3.1** - **3.8**), these cobalt complexes contain two ligands coordinated through the pyridyl nitrogen and amide oxygen to stabilise the 2+ oxidation state of the central cobalt ion.[1, 2, 5] Complex **3.9** contains two axial chloride ligands and complex **3.10** contains two axial aqua ligands resulting in a dicationic complex with two iodide counterions. Complex **3.10** shows that unlike thiocyanate, iodide is unable to displace the aqua ligands from the axial ligand positions. Both complexes adopt the *trans* geometry similar to complex **3.1** which contains the same picolinamide ligand and with

previous cobalt *bis*-picolinamide aqua complexes.[1-3] This supports the previous conclusion that the effect of ligand substituents on geometry is greater than the effect of the type of axial ligand. In addition, complex **3.9** was synthesised from a different cobalt starting material compared to all the other cobalt(II) *bis*-picolinamide complexes (cobalt chloride as opposed to cobalt nitrate) implying that the nature of the cobalt starting material has no bearing on the geometry of the final complex.

The synthesis of further cobalt *bis*-picolinamide complexes with imidazoles as the axial ligands using the same synthetic method as previously described was attempted; however these were mainly unsuccessful. A cobalt(II) *bis*-picolinamide thiocyanate imidazole complex was shown to form by mass spectrometry but could not be isolated (**Figure 3.14a**). In addition, the synthesis of monoanionic complexes containing one picolinamide ligand in the *N,O* coordination mode and the other picolinamide ligand in the *N,N* coordination mode was attempted. There was some evidence that the desired complex had formed by mass spectrometry, however this complex could not be isolated (**Figure 3.14b**). Further characterisation data is also required to determine whether the desired complex was formed, as opposed to the cobalt *bis*-picolinamide thiocyanate complex containing both picolinamide ligands in either the *N,N* or *N,O* coordination mode.
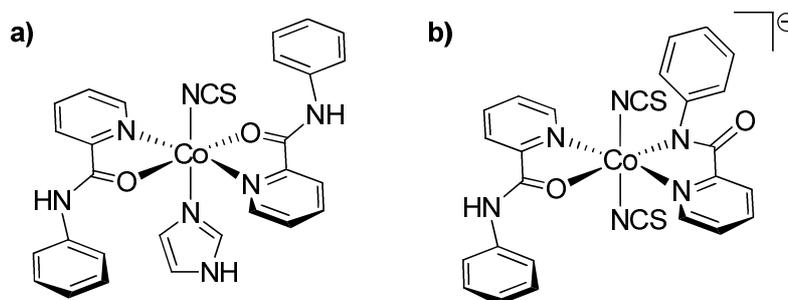
Figure 3.14: Cobalt *bis*-picolinamide complex with a) thiocyanate and imidazole ligands b) picolinamide ligands with mixed *N,O* and *N,N* coordination modes

3.2.1 X-Ray crystallography of cobalt *bis*-picolinamide complexes

Complexes were crystallised in a monoclinic (**3.9** and **3.10**) cell and structural solutions were performed in the *P* 2_1/c (**3.9** and **3.10**) space group with half (**3.9** and **3.10**) molecules in the asymmetric unit (**Figure 3.15**). Selected bond lengths and angles are stated in **Table 3.6** and **Table 3.7** respectively.

3.9   **3.10**
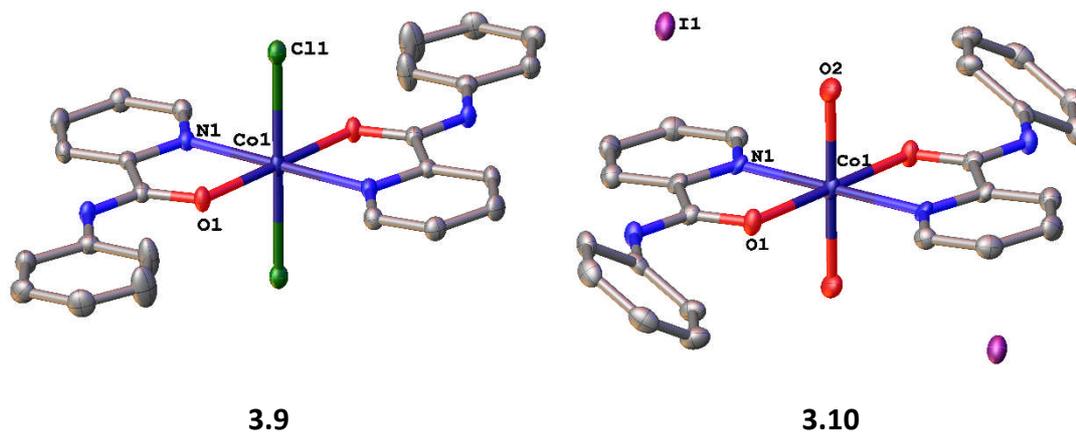
Figure 3.15: Molecular structures of complexes **3.9** and **3.10**. Displacement ellipsoids are at the 50 % probability level. Hydrogen atoms are omitted for clarity

Table 3.6: Selected bond lengths for complexes **3.9** and **3.10**. [a] = symmetry generated

Distance / Å	Complex	
	3.9	**3.10**
Co(1)-N(1)	2.0887(18)	2.094(3)
Co(1)-N(1)'	2.0887(18)[a]	2.094(3)
Co(1)-O(1)	2.0937(15)	2.046(2)
Co(1)-O(1)'	2.0937(15)[a]	2.046(2)
Co(1)-Cl(1)	2.4462(7)	-
Co(1)-Cl(1)'	2.4462(7)[a]	-
Co(1)-O(2)	-	2.121(3)
Co(1)-O(2)'	-	2.121(3)

Table 3.7: Selected bond angles for complexes **3.9** and **3.10**

	Angle / °			
	N(1)-Co(1)-O(1)	**O(1)-Co(1)-Cl(1)**	**O(1)-Co(1)-O(2)**	**O(1)-Co(1)-N(1)'**
3.9	77.42(7)	90.13(5)	-	102.58(7)
3.10	77.88(10)	-	91.71(10)	102.12(10)

Both complexes show a distorted octahedral geometry around the central cobalt atom with the relevant bond angles in the range 77.4 - 91.7°, deviating from the expected 90°. This distortion can be ascribed to the restrictions imposed by the bidentate picolinamide ligand. The cobalt-pyridyl nitrogen bond lengths are in the range 2.08 - 2.09 Å and the cobalt-oxygen bond lengths are in the range 2.04 - 2.09 Å. This is consistent with complex **3.1** which showed a Co-$N_{pyridyl}$ bond length of 2.11 Å and a Co-O_{amide} bond length of 2.10 Å. For complex **3.10**, the cobalt-aqua oxygen bond length is 2.12 Å which is consistent with reported cobalt *bis*-picolinamide aqua complexes which show Co-O_{aqua} bond lengths in the range 2.10 - 2.12 Å.[1-3] For complex **3.9** there is evidence from the crystal structure for an intramolecular hydrogen bond between the amide oxygen and the aryl *ortho*-CH group of the same ligand, however this hydrogen bond is not observed in complex **3.10** due to twisting of the aryl ring. Complex **3.9** also displays an intermolecular hydrogen bond between the chloride and the neighbouring amide NH group which is again absent in complex **3.10**. Other interactions observed include π-π stacking interactions between aryl rings and pyridyl rings in neighbouring molecules for complex **3.9** and bridging of complexes of **3.10** through the aqua ligands and iodide counterions (**Figure 3.16**). Overall, the planar nature of the picolinamide ligands in complex **3.9** permits similar intra- and intermolecular bonding interactions to complex **3.1** which also contains planar picolinamide ligands. The aryl rings of the picolinamide ligands in complex **3.10** are twisted and therefore the ligand is not planar, presumably this is due to the presence of the iodide counterion. This intra-ligand torsion alters the nature of the intermolecular interactions and thus complex **3.10** does not display any π-π stacking interactions.

a)

b)
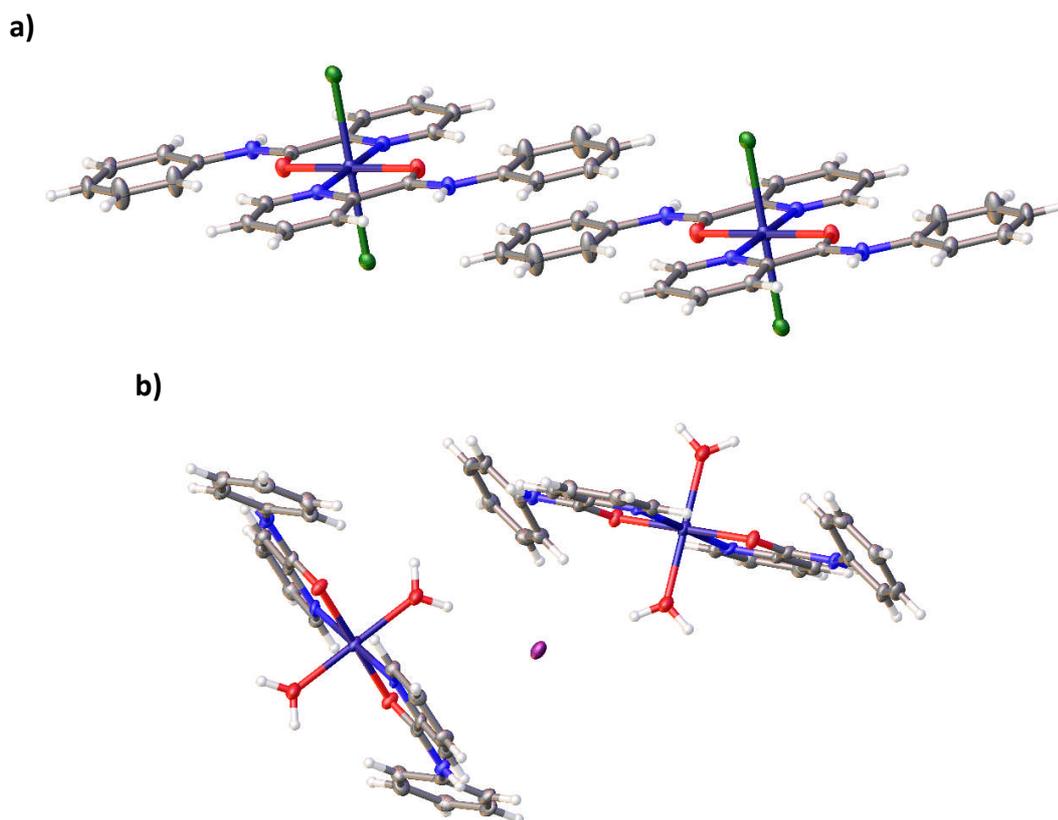
Figure 3.16: Intermolecular interactions a) π-π stacking b) bridging of complexes through aqua ligands and iodide counterion

3.3 Conclusions

A series of novel high spin cobalt(II) *bis*-picolinamide complexes has been synthesised with the two picolinamide ligands coordinating in the *N,O* mode and the two axial ligands either thiocyanate, chloride or aqua. The substituent on the aryl ring of the ligands determines the *trans* or *cis* geometry of the final complexes. In general, no substituents or *meta* substituents lead to the *trans* geometry, whereas *ortho* or *para* substituents bestow the *cis* geometry. Complexes show a distorted octahedral structure with bond lengths in keeping with a cobalt(II) species. Intramolecular hydrogen bonds are observed for *trans* complexes but not for *cis* complexes. Intermolecular hydrogen bonds and π-π stacking interactions are observed between neighbouring molecules. *Trans* complexes are hypothesised to be the thermodynamic product of the reaction. The efficacy of the complexes as anti-cancer, anti-bacterial and anti-fungal agents will be discussed in **Chapter 5**.

3.4 References

1.   G. V. Tsintsadze, R. A. Kiguradze, A. N. Shnulin and K. S. Mamedov, *Journal of Structural Chemistry*, 1986, **27**, 272-278.

2.   J. Xue, X. Hua, L. Yang, W. Li, Y. Xu, G. Zhao, G. Zhang, L. Liu, K. Liu, J. e. Chen and J. Wu, *Journal of Molecular Structure*, 2014, **1059**, 108-117.

3.   O. Z. Yesilel, H. Olmez, O. O. Yilan, H. Pasaoglu and O. Buyukgungor, *Zeitschrift Fur Naturforschung Section B- A Journal of Chemical Sciences*, 2006, **61**, 1094-1100.

4.   M. Dakovic, M. Vinkovic, S. Roca, Z. Popovic, I. Vickovic, D. Vikic-Topic, J. Lukac, N. Dakovic and Z. Kusic, *Journal of Coordination Chemistry*, 2012, **65**, 1017-1032.

5.   M. Dakovic, D. Vila-Vicosa, M. J. Calhorda and Z. Popovic, *CrystEngComm*, 2011, **13**, 5863-5871.

6.   G. Lanza, I. L. Fragala and T. J. Marks, *Journal of the American Chemical Society*, 2000, **122**, 12764-12777.

7.   M. J. Deetz, J. E. Fahey and B. D. Smith, *Journal of Physical Organic Chemistry*, 2001, **14**, 463-467.

8.   J. S. Laursen, J. Engel-Andreasen, P. Fristrup, P. Harris and C. A. Olsen, *Journal of the American Chemical Society*, 2013, **135**, 2835-2844.

9.   A. Ali, D. Bansal and R. Gupta, *Journal of Chemical Sciences*, 2014, **126**, 1535-1546.

10.   A. Mishra, A. Ali, S. Upreti and R. Gupta, *Inorganic Chemistry*, 2008, **47**, 154-161.

11.   J. Y. Qi, H. X. Ma, X. J. Li, Z. Y. Zhou, M. C. K. Choi, A. S. C. Chan and Q. Y. Yang, *Chemical Communications*, 2003, 1294-1295.

12.   M. Ray, D. Ghosh, Z. Shirin and R. Mukherjee, *Inorganic Chemistry*, 1997, **36**, 3568-3572.

13.   C. E. Housecroft and A. G. Sharpe, *Inorganic Chemistry*, Fourth edn., Pearson Ltd., 2012.

14.   H. A. Goodwin, *Spin Crossover in Transition Metal Compounds II*, 2004, **234**, 23-47.

15.   M. A. Halcrow, *Chemical Communications*, 2013, **49**, 10890-10892.

16.   S. Hayami, Y. Komatsu, T. Shimizu, H. Kamihata and Y. N. Lee, *Coordination Chemistry Reviews*, 2011, **255**, 1981-1990.

17.   R. G. Miller, S. Narayanaswamy, J. L. Tallon and S. Brooker, *New Journal of Chemistry*, 2014, **38**, 1932-1941.

18.   P. Pietrzyk, M. Srebro, M. Radon, Z. Sojka and A. Michalak, *Journal of Physical Chemistry A*, 2011, **115**, 2316-2324.

19.   R. A. Taylor, A. J. Lough and M. T. Lemaire, *Journal of Materials Chemistry C*, 2016, **4**, 455-459.

20.   D. F. Evans, *Journal of the Chemical Society*, 1959, 2003-2005.

21.   D. Robbes, *Sensors and Actuators A-Physical*, 2006, **129**, 86-93.

Chapter 4

Synthesis and characterisation of cobalt (III) mixed ligand complexes

4. Cobalt (III) mixed ligand complexes

There are a number of examples of cobalt mixed ligand complexes in the literature, however none of these complexes contain picolinamide ligands. In terms of anti-cancer cobalt mixed ligand complexes, the majority of these contain a known cytotoxic ligand with the cobalt and additional ligands present either to act as a chaperone or for tunability purposes, to allow the more selective targeting of cancer cells. Cobalt(II) conjugates containing pyridyl based ligands, e.g. pyridine, bipyridine and phenanthroline, and nonsteroidal anti-inflammatory drugs e.g. naproxen, mefenamic acid and tolfenamic acid have shown strong DNA binding properties and high antioxidant activities but have not been investigated for cytotoxicity (**Figure 4.1a**).[1-3] A cobalt(III) cyclam complex with naproxen was found to be toxic to both cancer cells and cancer stem cells.[4] Cobalt(III) mixed ligand complexes with acetylacetonate and nitrogen mustard ligands have shown to be cytototoxic with improved targeting of cancerous cells over non-cancerous cells. This targeting is due to hypoxia selectivity, as in a hypoxic environment the central cobalt(III) is reduced to cobalt(II) with the concurrent loss of the nitrogen mustard ligand which is a known therapeutic agent (**Figure 4.1b**).[5, 6] Cobalt(III) mixed ligand complexes with *tris*(2-methylpyridyl)amine (tpa) with marimastat, a matrix metalloproteinase, was found to suppress tumour growth. Complexation of marimastat through the hydroxamic acid moiety, thought to be the cause of low bioavailability due to binding to other biomolecules, imparted hypoxic selectivity allowing the specific targeting of cancer cells (**Figure 4.1c**).[7, 8] Aside from tpa, cyclen ligands are commonly used in the preparation of cobalt(III) chaperones.[9, 10]
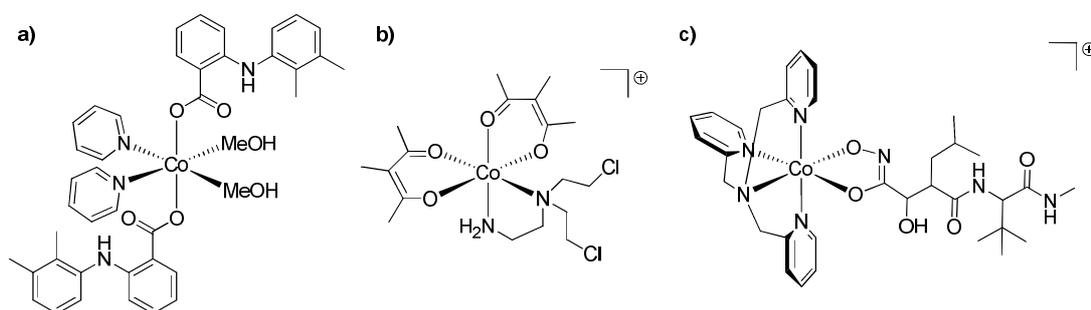
Figure 4.1: The structures of a) cobalt-mefenamic acid conjugate b) cobalt acac nitrogen mustard c) cobalt tpa marimastat

4.1 β-Diketonate and ferrocenyl β-diketonate ligands

Ferrocene is a metallocene consisting of a central iron(II) ion "sandwiched" between two cyclopentadiene (Cp) rings. Ferrocene has good aqueous solubility, with highly tunable properties and is easily derivatised, leading to the widespread investigation of ferrocene bioconjugates and ferrocenyl drug-derivatives.[11] To date, the only ferrocenyl drug derivative to have entered the clinic is the anti-malarial drug ferroquine (**Figure 4.2a**).[12] In terms of chemotherapy, ferrocifen: the ferrocene adduct of hydroxytamoxifen (the active metabolite of the breast cancer drug tamoxifen) (**Figure 4.2b**), showed improved bioavailability and activity against both hormone-dependent and hormone-independent tumours.[13, 14]
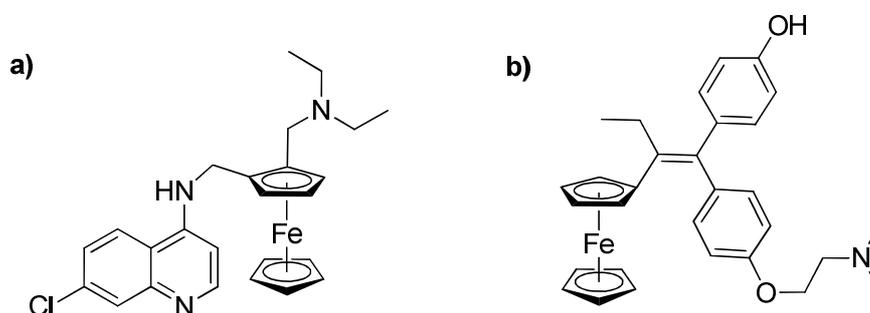
Figure 4.2: The structures of a) ferroquine b) ferrocifen

The successes of ferrocenyl derivatives in clinical research has led to the investigation of a number of ferrocenyl derivatives of compounds which can act as ligands for metal complexes. For example, β-diketonate ligands are non-cytotoxic but ferrocenyl β-diketonate ligands have shown promising cytotoxic activity, these are currently under investigation within the McGowan group.[15] Within the series of ferrocenyl β-diketonate ligands, higher reduction potentials and lower pK_a values are associated with greater toxicity.[16] Metal complexes with these ferrocenyl β-diketonate ligands have also proven cytotoxic, but less cytotoxic than the free ligand.[17]

β-Diketonate ligands were synthesised *via* the Claisen condensation reaction of ethyl acetate with the respective acetophenone in presence of sodium ethoxide (**Scheme 4.1a**).[18] All ligands have been synthesised previously in the literature and are not discussed further.[19-21] Ferrocenyl β-diketonate ligands were synthesised *via* the Claisen condensation reaction of acetyl ferrocene with the respective benzoate or acetate in the presence of sodium ethoxide (**Scheme 4.1b**).[16, 22] All ferrocenyl

β-diketonate ligands were synthesised by Matthew Allison at The University of Leeds.
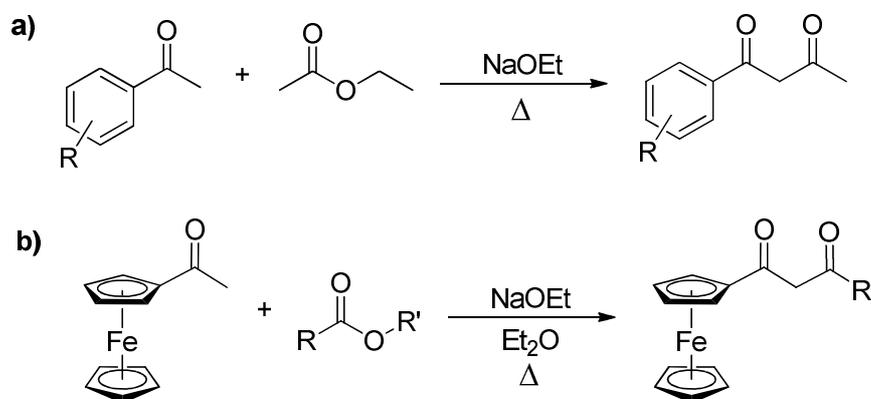
Scheme 4.1: Synthetic routes for β-diketonate ligands

The ^{1}H NMR spectra of the β-diketonate and ferrocenyl β-diketonate ligands show a characteristic methine proton singlet peak between 5.63 - 7.01 ppm depending upon the nature of the substitution on the ligand; this signal is the most characteristic for ligand formation.

4.2 Cobalt *bis*-picolinamide β-diketonate complexes

The cobalt picolinamide complexes synthesised within this section are shown in **Figure 4.3**. Complexes were synthesised *via* the complexation reaction of picolinamide and β-diketonate ligands with cobalt carbonate in the presence of hydrogen peroxide, similar to the synthesis used for the preparation of cobalt β-diketonate complexes (**Scheme 4.2**).[23-25] Complex **4.1** was prepared and partially characterised by Bilal Saloo at The University of Leeds following the procedure determined by the author.
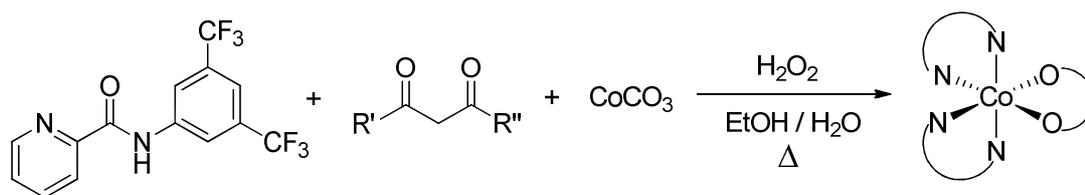
Scheme 4.2: Synthetic route for cobalt picolinamide β-diketonate complexes in this section

R =   H   (4.1)   R =   H   (4.3)   3-Me   (4.7)

       Ph   (4.2)   3-F   (4.4)   4-Me   (4.8)

                                   4-F   (4.5)   4-OMe   (4.9)

                                   4-CF$_3$ (4.6)
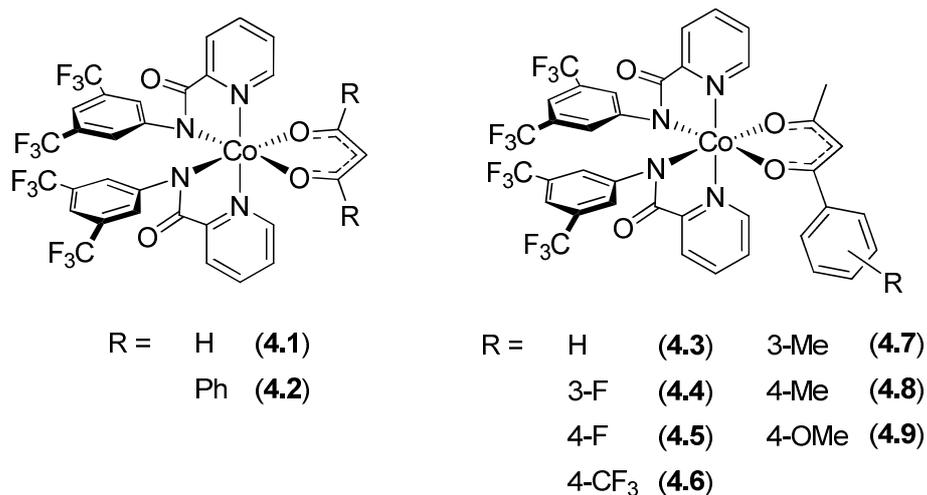
Figure 4.3: Cobalt picolinamide complexes synthesised within this section

In this synthesis, cobalt is oxidised from Co(II) in the metal carbonate precursor to Co(III) in the metal complex which then permits the complexation of three ligands to the metal centre. The β-diketonate ligand coordinates through the two keto oxygen atoms and the two picolinamide ligands coordinate through the pyridyl nitrogen and amide nitrogen, acting as monoanionic donors to stabilise the 3+ oxidation state of the central cobalt ion. This *N,N* coordination of the picolinamide ligands is ensured with the use of an electron withdrawing group substituent on the aryl ring and the use of a 2:1 ethanol-water mixture as the reaction solvent. The same picolinamide ligand is used throughout and the β-diketonate ligand varied; the 3,5-di(trifluoromethyl) picolinamide ligand (**1.14**) was selected as the analogous cobalt(III) *tris*-picolinamide complex with this ligand was found to be cytotoxic (**Chapter 5**). The presence of hydrogen peroxide in the reaction is responsible for the oxidation of cobalt in an aerobic atmosphere. Cobalt may also be oxidised with bases such as triethylamine or sodium hydrogen carbonate, however these result in poorer conversion of the metal precursor to the final complex or the formation of a greater percentage of side products.

The formation of complexes **4.1** - **4.9**, with two picolinamide ligands and one β-diketonate ligand, is permitted through stoichiometric control of the reaction. Despite this a number of side products are also formed resulting in lower yields. The two major side products formed are the cobalt(III) *tris*-picolinamide complex and the cobalt(III) picolinamide β-diketonate complex with one picolinamide ligand and two β-diketonate ligands, as shown by thin layer chromatography (TLC) and mass spectrometry.

The synthesis was successful with the majority of β-diketonate ligands apart from curcumin (**Figure 4.4**). The cobalt picolinamide curcumin complex was shown to form by mass spectrometry but could not be isolated. Curcumin is a natural product present in turmeric which has been found to have anti-tumourigenic, anti-angiogenic and anti-inflammatory properties and is thus under investigation as a potential anti-cancer drug.[26-29] However, curcumin undergoes rapid metabolism and is poorly soluble in water (especially at physiological pH) leading to poor bioavailability.[30, 31] Various methods are under investigation to improve bioavailability, protect against degradation and target cancer cells including the use of chaperones and nanoparticles as delivery mechanisms. A few cobalt curcumin complexes have been evaluated for their potential to act as curcumin chaperones with promising results.[32-38]
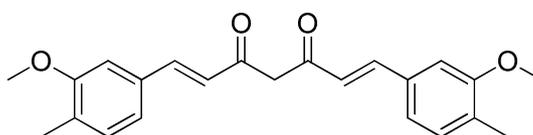
Figure 4.4: The structure of curcumin

4.2.1 Characterisation of cobalt picolinamide β-diketonate complexes

The cobalt(III) picolinamide β-diketonate complexes are low spin d^6 and thus diamagnetic. The ^{1}H NMR spectra of these complexes are all very similar with the pyridyl protons (8.19 - 7.13 ppm) typically having higher chemical shifts than the aryl protons of both the picolinamide and β-diketonate ligands (7.90 - 6.91 ppm) and the methine proton in the region 6.79 - 5.43 ppm. All pyridyl and aryl protons from both ligand types are shifted upfield in the complex compared to in the free ligand; the methine proton also undergoes a slight upfield shift. Examples of the ^{1}H NMR chemical shifts for complex **4.8** compared to the ligands are shown in **Figure 4.5** and **Table 4.1**. Two distinct or overlapping sets of signals for the picolinamide protons are seen in the final complex as the two ligands are inequivalent due to the slightly distorted octahedral geometry of the complex.
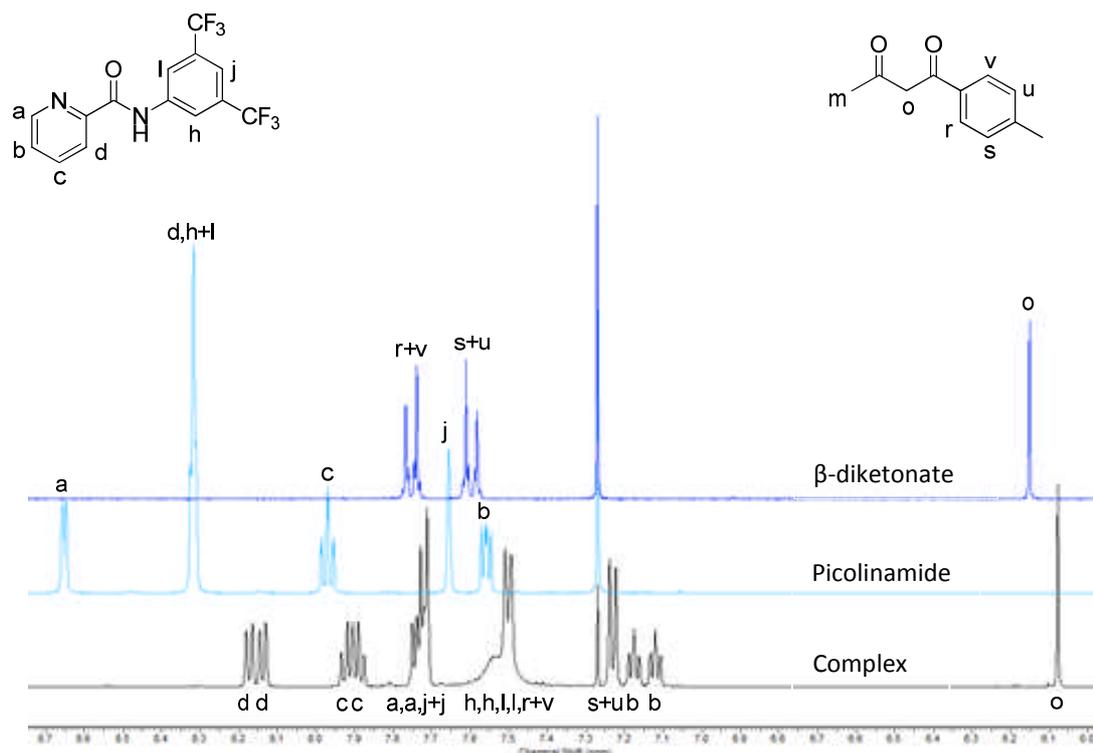
Figure 4.5: ^{1}H NMR spectra of complex **4.8** and the constituent picolinamide and β-diketonate ligands

Table 4.1: ^{1}H NMR chemical shifts for complex **4.8** and the constituent picolinamide and β-diketonate ligands

Proton environment	^{1}H NMR chemical shift / ppm	
	Ligand	Complex
Pyridyl	8.66	8.16
	8.30	7.91
	7.97	7.73
	7.48	7.15
Aryl	8.32	7.73
(picolinamide)	7.68	7.52
Aryl	7.76	7.52
(β-diketonate)	7.60	7.24
Methine	6.15	6.08

The IR spectra of complexes **4.1** - **4.9** all display a weak aryl CH stretch at 3080 - 3100 cm^{-1}, a medium intensity CO (picolinamide) stretch at 1635 - 1640 cm^{-1} and a medium intensity CO (β-diketonate) stretch at 1600 - 1605 cm^{-1}. There are also several bands of varying intensities in the region 500 - 1600 cm^{-1}. The disappearance of the broad NH stretches (3250 - 3400 cm^{-1}) is characteristic of complex formation with the picolinamide ligand adopting the *N,N* coordination mode. The CO stretches for the picolinamide and β-diketonate ligand are shifted to a lower wavenumber in the complex compared to the free ligand as is expected due to the weakening of the CO bond following coordination of the ligand to the metal centre.[39, 40]

The UV/vis spectrum of complex **4.3** is shown in **Figure 4.6**. The spectrum shows an intense ligand based absorbance (π-π*) at around 200 nm with three less intense charge-transfer bounds at around 265, 290 and 360 nm. No d-d transitions are seen in the 400 - 800 nm region.[39, 40]
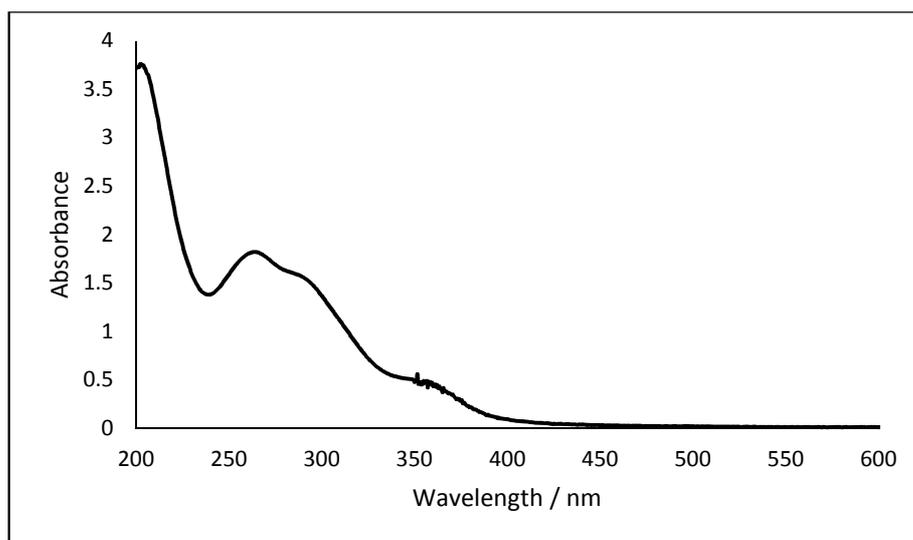
Figure 4.6: UV/vis spectrum of complex **4.3**

4.2.2 X-Ray crystallography of cobalt picolinamide β-diketonate complexes

Complexes crystallised in either a monoclinic (**4.3**, **4.4**, **4.6**, **4.8**, **4.9**), triclinic (**4.2**) or tetragonal (**4.1**) cell and structural solutions were performed in the *P* 2$_1$/n (**4.3**), *P* 2$_1$/c (**4.4**, **4.6**), *P* $\overline{1}$ (**4.2**), *C* c (**4.8**, **4.9**) or *I* 4$_1$cd (**4.1**) space group with half (**4.1**), one (**4.2**, **4.4**, **4.6**, **4.8**, **4.9**) or two (**4.3**) molecules in the asymmetric unit (**Figure 4.7**). Selected bond lengths and angles are stated in **Table 4.2** and **Table 4.3** respectively.
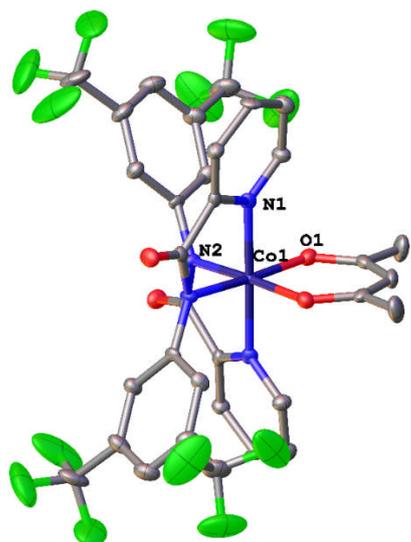
4.1
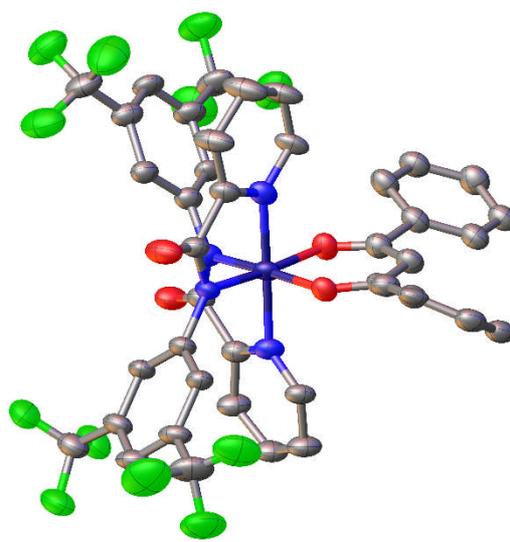
4.2
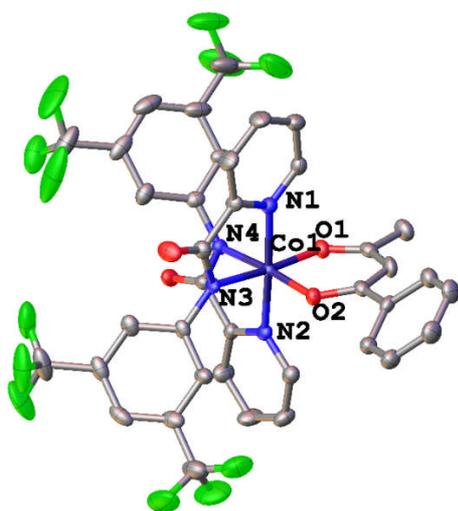
4.3
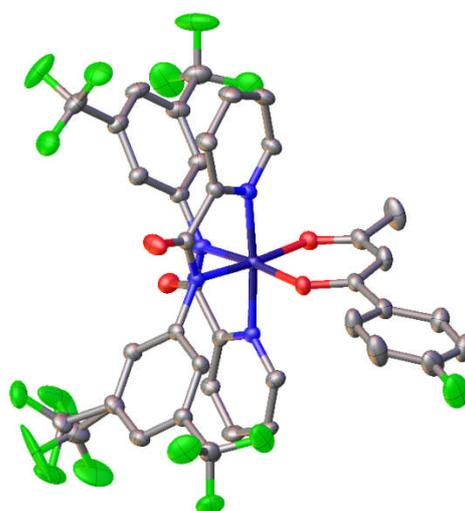
4.4
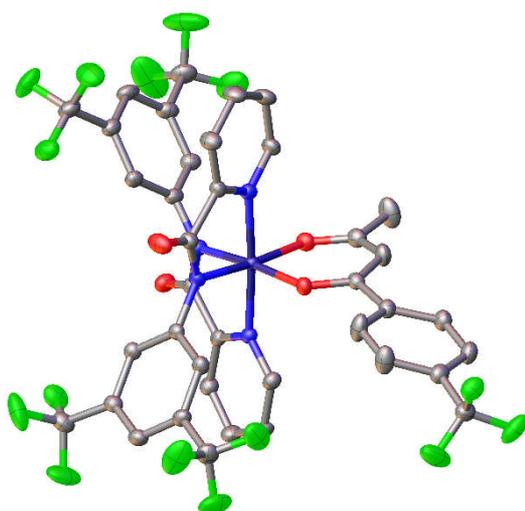
4.6
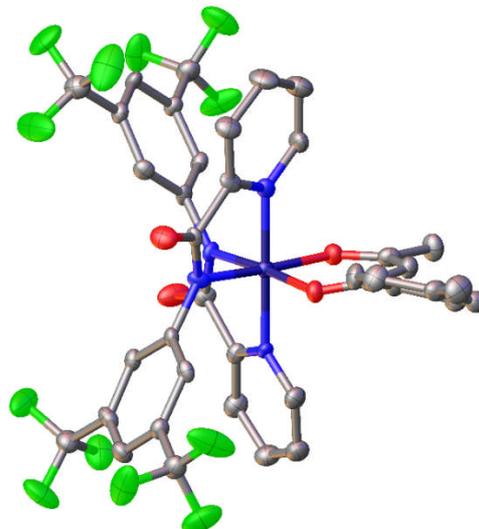
4.8
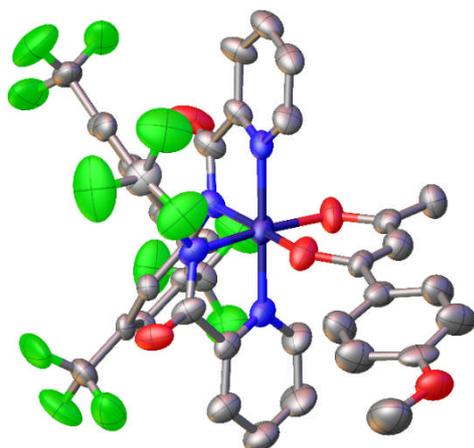
4.9

Figure 4.7: Molecular structures of complexes **4.1** - **4.9**. Displacement ellipsoids are at the 50 % probability level. Hydrogen atoms and co-crystallising solvents are omitted for clarity. For clarity, only complexes **4.1** and **4.3** are labelled, the remaining complexes follow the same atom numbering scheme

Table 4.2: Selected bond lengths for complexes **4.1** - **4.9**. [a] = symmetry generated

	Distance / Å					
	Co(1)-N(1)	Co(1)-N(2)	Co(1)-N(3)	Co(1)-N(4)	Co(1)-O(1)	Co(1)-O(2)
4.1	1.939(5)	1.939(5)[a]	1.938(6)	1.938(6)[a]	1.903(5)	1.903(5)[a]
4.2	1.944(15)	1.898(17)	1.947(14)	1.942(12)	1.909(15)	1.908(9)
4.3	1.923(4)	1.928(4)	1.926(4)	1.941(4)	1.903(3)	1.908(3)
4.4	1.9340(16)	1.9353(16)	1.9322(16)	1.9330(17)	1.9030(14)	1.9043(14)
4.6	1.9311(17)	1.9381(17)	1.9291(17)	1.9317(18)	1.9041(15)	1.9063(15)
4.8	1.928(6)	1.943(6)	1.937(6)	1.947(5)	1.906(5)	1.922(5)
4.9	1.958(14)	1.910(14)	1.947(11)	1.924(12)	1.873(11)	1.947(10)

Table 4.3: Selected bond angles for complexes **4.1** - **4.9**

	Angle / °		
	N(1)-Co(1)-N(3)	**N(3)-Co(1)-O(2)**	**O(1)-Co(1)-O(2)**
4.1	83.5(2)	85.67(19)	95.0(3)
4.2	83.8(7)	87.3(5)	93.6(5)
4.3	83.23(15)	87.56(15)	94.41(14)
4.4	82.55(7)	86.16(6)	94.35(6)
4.6	82.61(7)	86.74(7)	93.97(6)
4.8	82.6(2)	89.5(2)	93.9(2)
4.9	82.5(5)	90.7(5)	95.7(5)

All complexes show a distorted octahedral geometry around the central cobalt atom with the relevant bond angles in the range 82.5 - 95.7°, deviating from the expected 90° (**Table 4.3**). This distortion can be ascribed to the restrictions imposed by the bidentate picolinamide and β-diketonate ligand. The cobalt-pyridyl nitrogen bond lengths are in the range 1.94 - 1.96 Å, the cobalt-amide nitrogen bond lengths are in the range 1.92 - 1.95 Å and the cobalt-oxygen bond lengths in the range 1.87 - 1.95 Å. This is consistent with related low spin Co(III) compounds which have reported Co-$N_{pyridyl}$ bond lengths of 1.91 - 1.96 Å and Co-N_{amide} bond lengths of 1.90 - 1.99 Å.[41-44] These are also shorter than in related Co(II) compounds which have reported Co-$N_{pyridyl}$ bond lengths of 2.07 - 2.15 Å.[45-47] The Co-$O_{diketonate}$ bond lengths are also consistent with literature values which are in the range 1.84 - 1.91 Å.[48-50] There is evidence from the crystal structure for long range intermolecular hydrogen bonds, with bond distances in the range 3.12 - 3.38 Å. These hydrogen bonds tend to be either between the amide oxygen and a neighbouring pyridyl or aryl CH group or between the trifluoromethyl group and a neighbouring aryl CH group (**Figure 4.8**).
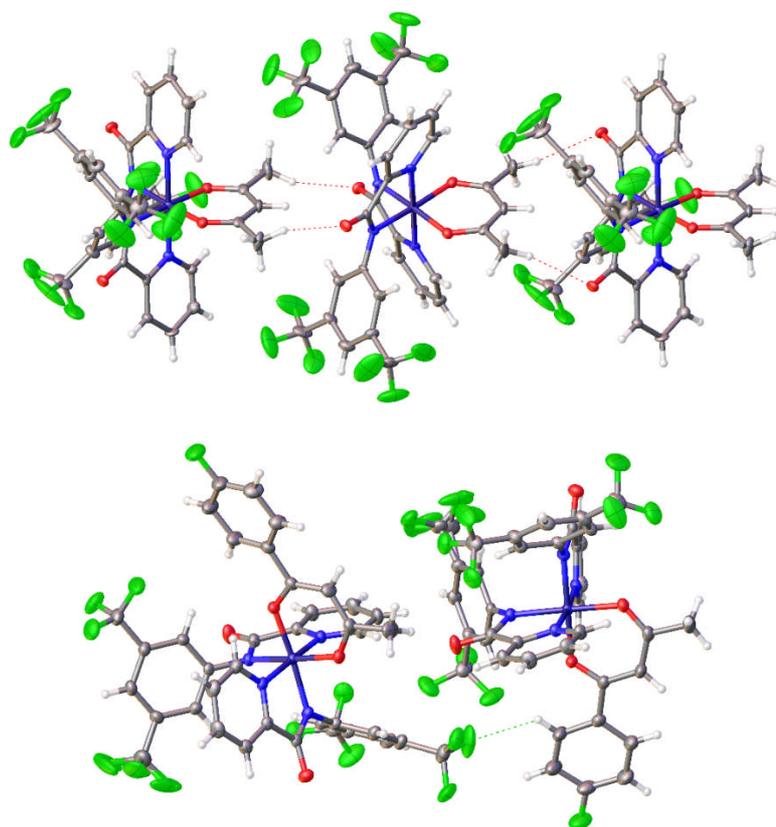
Figure 4.8: Intermolecular hydrogen bonding interactions between complexes

4.3 Cobalt *bis*-picolinamide ferrocenyl β-diketonate complexes

The cobalt picolinamide complexes synthesised within this section are shown in **Figure 4.9**. Complexes were synthesised *via* the complexation reaction of picolinamide and β-diketonate ligands with cobalt carbonate in the presence of hydrogen peroxide, similar to the synthesis in the previous section (**Scheme 4.3**).
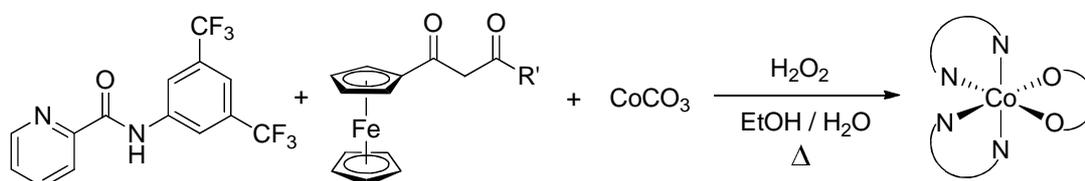
Scheme 4.3: Synthetic route for cobalt picolinamide ferrocenyl β-diketonate complexes in this section

R =   CHF$_2$ (**4.10**)   R =   H   (**4.13**)   3-Me   (**4.17**)

      CF$_3$ (**4.11**)   3-F   (**4.14**)   4-Me   (**4.18**)

      Me   (**4.12**)   4-F   (**4.15**)   3,5-diMe   (**4.19**)

                                      3,5-diF (**4.16**)
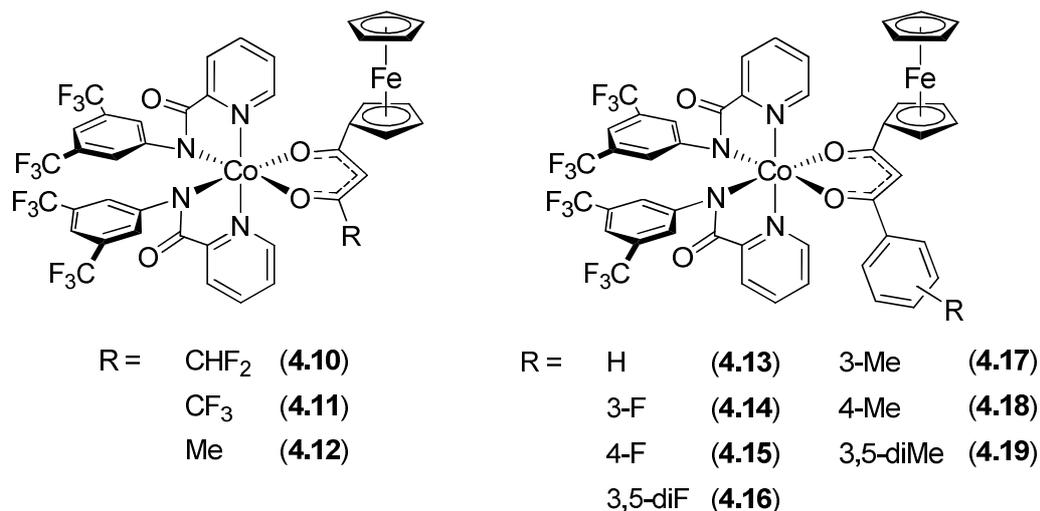
Figure 4.9: Cobalt picolinamide complexes synthesised within this section

As in the previous synthesis, cobalt is oxidised from Co(II) to Co(III) which then permits the complexation of three ligands to the metal centre. The β-diketonate ligand coordinates through the two keto oxygen atoms and the two picolinamide ligands coordinate through the pyridyl nitrogen and amide nitrogen. This *N,N* coordination of the picolinamide ligands is ensured with the use of an electron withdrawing group substituent on the aryl ring and the use of a 2:1 ethanol-water mixture as the reaction solvent.

The formation of the desired complex with two picolinamide ligands and one ferrocenyl β-diketonate ligand is permitted through stoichiometric control of the reaction. As for the previous synthesis, a number of side products are also formed, however these are more easily separated resulting in improved yields compared to the cobalt(III) picolinamide β-diketonate complexes. The two major side products formed are the cobalt(III) *tris*-picolinamide complex and the cobalt(III) picolinamide ferrocenyl β-diketonate complex with one picolinamide ligand and two ferrocenyl β-diketonate ligands, as shown by TLC and mass spectrometry.

4.3.1 Characterisation of cobalt picolinamide ferrocenyl β-diketonate complexes

The cobalt(III) picolinamide ferrocenyl β-diketonate complexes are low spin d^6 and thus diamagnetic. The ^{1}H NMR spectra of these complexes are all very similar to each other and to the cobalt complexes containing the non-ferrocenyl β-diketonate ligand. The pyridyl protons (8.34 - 7.13 ppm) typically have higher chemical shifts

than the aryl protons of both the picolinamide and β-diketonate ligands (7.85 - 6.99 ppm), with the methine proton in the region 6.29 - 5.61 ppm and the ferrocenyl protons in the region 4.99 - 2.10 ppm. As for the equivalent non-ferrocenyl complexes, all pyridyl and aryl protons from both ligand types are shifted upfield in the complex compared to in the free ligand; the methine proton and ferrocenyl protons also undergo an upfield shift. Examples of the ^{1}H NMR chemical shifts for complex **4.16** compared to the ligands are shown in **Figure 4.10** and **Table 4.4**. Two distinct or overlapping sets of signals for the picolinamide protons are seen in the final complex as the two ligands are inequivalent due to the slightly distorted octahedral geometry of the complex. The ferrocenyl protons (substituted Cp ring) also split from two to four peaks showing the breaking of symmetry and inequivalence of these protons upon complexation.
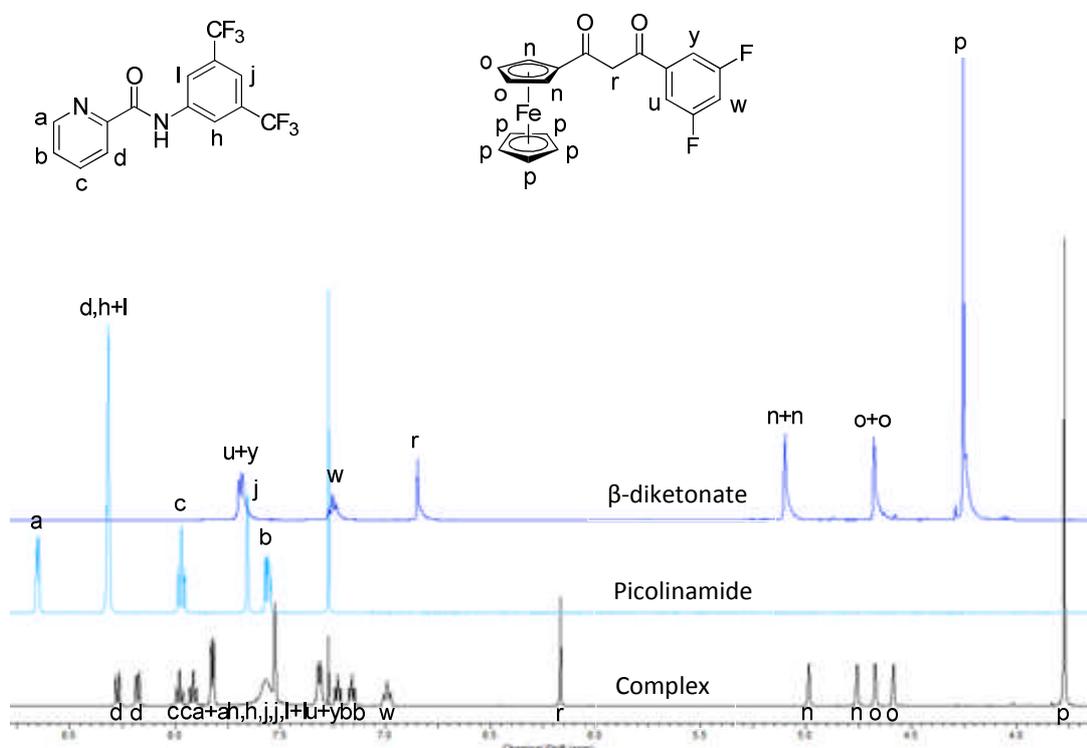
Figure 4.10: ^{1}H NMR spectra of complex **4.16** and the constituent picolinamide and ferrocenyl β-diketonate ligands

Table 4.4: ^{1}H NMR chemical shifts for complex **4.16** and the constituent picolinamide and β-diketonate ligands

Proton environment	^{1}H NMR chemical shift / ppm	
	Ligand	Complex
Pyridyl	8.66	8.28 and 8.18
	8.30	7.98 and 7.91
	7.97	7.83
	7.48	7.23 and 7.16
Aryl	8.32	7.57
(picolinamide)	7.68	7.53
Aryl	7.56	7.32
(β-diketonate)	7.12	6.99
Methine	6.72	6.17
Ferrocenyl	4.97	4.99 and 4.76
	4.55	4.67 and 4.59
	4.13	3.77

The IR spectra of complexes **4.10** - **4.19** all display a weak aryl CH stretch at 3050 - 3090 cm^{-1}, a medium intensity CO (picolinamide) stretch at 1635 - 1640 cm^{-1} and a medium intensity CO (ferrocenyl β-diketonate) stretch at 1600 - 1605 cm^{-1}. There are also several bands of varying intensities in the region 500 - 1600 cm^{-1}. The IR spectra of these complexes are similar to the equivalent non-ferrocenyl complexes (**4.1** - **4.9**). The disappearance of the broad NH stretches (3250 - 3400 cm^{-1}) is characteristic of complex formation with the picolinamide ligand adopting the *N,N* coordination mode. The CO stretches of both the picolinamide and ferrocenyl β-diketonate ligand are shifted to a lower wavenumber in the complex compared to the free ligand due to the weakening of the CO bond following coordination of the ligand to the metal centre.[16, 51]

The UV/vis spectrum of complex **4.13** is shown in **Figure 4.11**. The spectrum shows an intense ligand based absorbance (π-π*) at around 200 nm with three less intense charge-transfer bounds at around 265, 300 and 370 nm and finally a weak d-d transition at around 515 nm. The absorbance bands are similar to in the

equivalent non-ferrocenyl complex **4.3** (**Figure 4.6**) aside from the d-d transition. Complex **4.3** shows no d-d transition whereas complex **4.13** shows a clear absorbance band, therefore this d-d transition can be assigned to the iron(II) of the ferrocenyl β-diketonate rather than the central cobalt(III) ion.[16, 51]
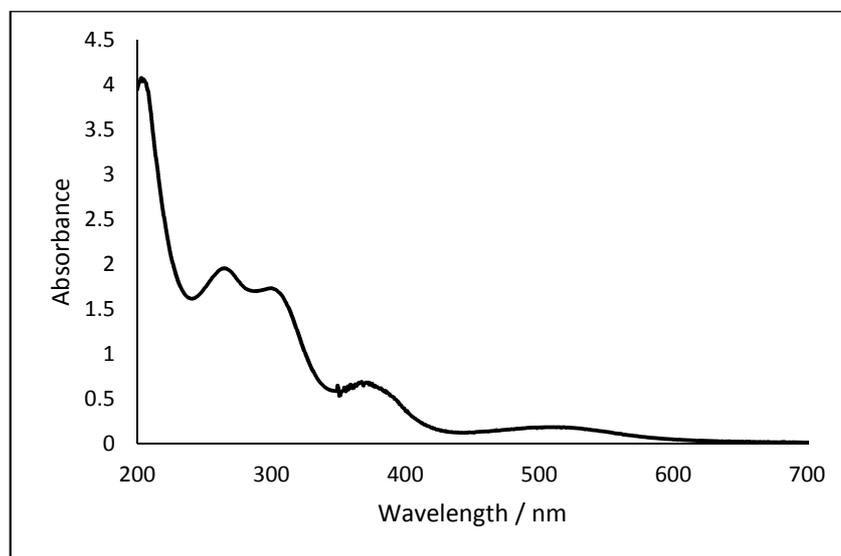
Figure 4.11: UV/vis spectrum of complex **4.13**

4.3.2 X-Ray crystallography of cobalt picolinamide ferrocenyl β-diketonate complexes

Complexes crystallised in either a monoclinic (**4.13**, **4.14**, **4.15**, **4.16**, **4.18**) or orthorhombic (**4.11**, **4.17**, **4.19**) cell and structural solutions were performed in the *P* 2₁ (**4.16**), *P* 2₁/c (**4.13**, **4.18**), *P* 2/c (**4.14**), *P* na2₁ (**4.11**, **4.17**, **4.19**) or *I* a (**4.15**) space group with half (**4.14**), one (**4.11**, **4.14**, **4.17**, **4.18**, **4.19**) or two (**4.13, 4.16**) molecules in the asymmetric unit (**Figure 4.12**). Selected bond lengths and angles are stated in **Table 4.5** and **Table 4.6** respectively.
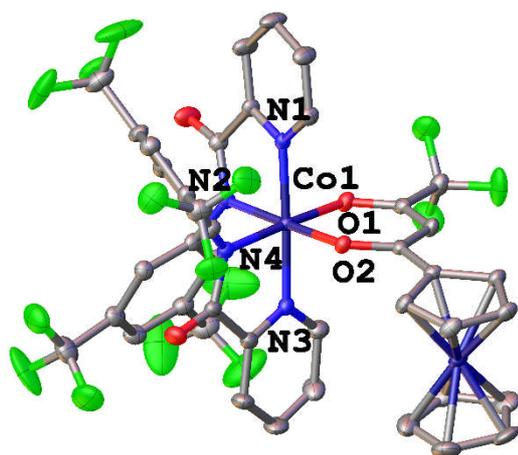
4.11

4.13

4.14

4.15

4.16

4.17

4.18   **4.19**
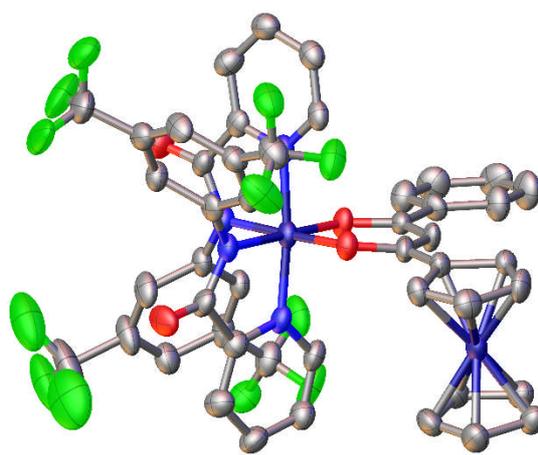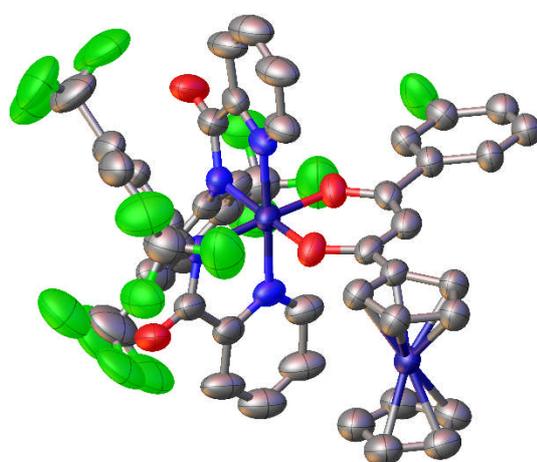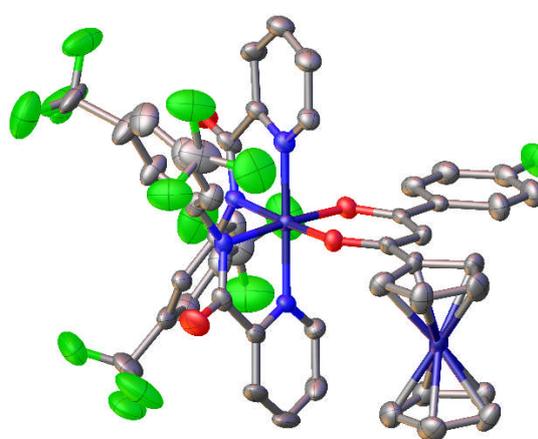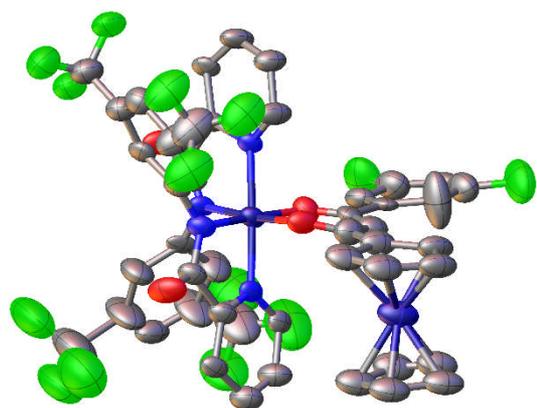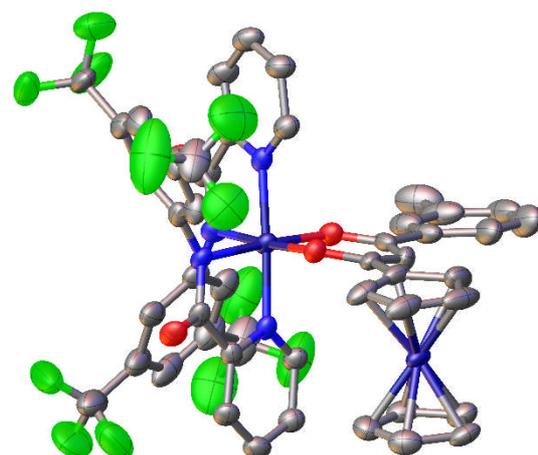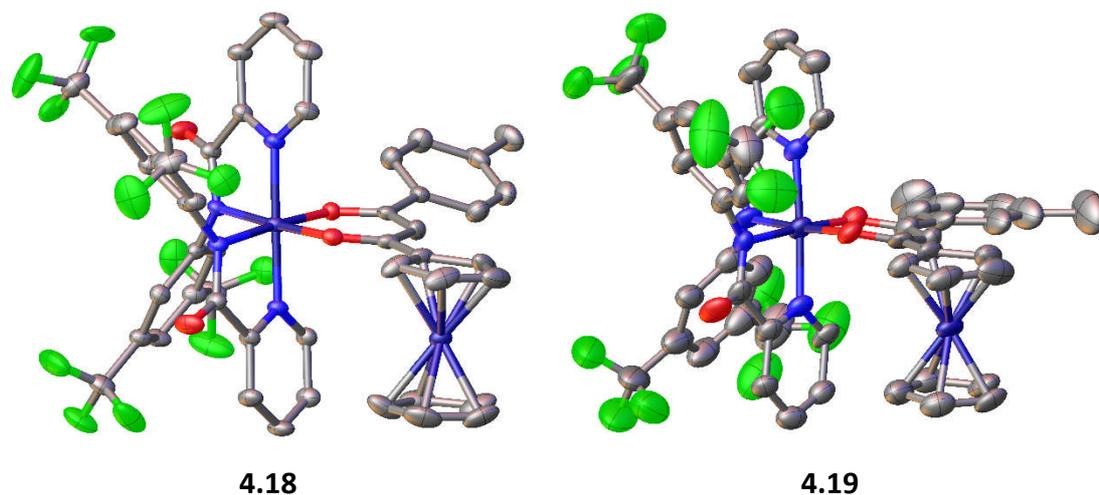
Figure 4.12: Molecular structures of complexes **4.11** - **4.19**. Displacement ellipsoids are at the 50 % probability level. Hydrogen atoms and co-crystallising solvents are omitted for clarity. For clarity, only complex **4.11** is labelled, the remaining complexes follow the same atom numbering scheme

Table 4.5: Selected bond lengths for complexes **4.11** - **4.19**. [a] = symmetry generated

	Distance / Å					
	Co(1)-N(1)	Co(1)-N(2)	Co(1)-N(3)	Co(1)-N(4)	Co(1)-O(1)	Co(1)-O(2)
4.11	1.916(3)	1.920(3)	1.932(4)	1.922(4)	1.917(3)	1.914(3)
4.13	1.919(7)	1.935(7)	1.923(7)	1.928(6)	1.904(5)	1.893(6)
4.14	1.938(7)	1.933(8)	1.938(7)[a]	1.933(8)[a]	1.896(7)	1.896(7)[a]
4.15	1.935(7)	1.936(7)	1.939(7)	1.925(7)	1.878(6)	1.890(5)
4.16	1.92(3)	1.92(3)	1.91(3)	1.90(3)	1.90(3)	1.91(2)
4.17	1.927(6)	1.915(6)	1.931(6)	1.944(5)	1.917(5)	1.901(5)
4.18	1.925(2)	1.930(3)	1.919(2)	1.933(3)	1.886(2)	1.900(2)
4.19	1.928(8)	1.906(9)	1.933(8)	1.938(9)	1.881(8)	1.909(8)

Table 4.6: Selected bond angles for complexes **4.11** - **4.19**

	Angle / °		
	N(1)-Co(1)-N(2)	**N(2)-Co(1)-O(1)**	**O(1)-Co(1)-O(2)**
4.11	83.13(15)	87.56(16)	94.95(14)
4.13	83.2(3)	87.4(3)	95.1(2)
4.14	83.7(3)	86.6(3)	95.8(4)
4.15	83.2(3)	87.0(3)	94.9(2)
4.16	82.8(12)	88.3(13)	92.2(12)
4.17	82.5(2)	87.5(2)	94.3(2)
4.18	82.77(11)	85.89(10)	94.89(10)
4.19	83.6(4)	86.6(4)	95.4(3)

All complexes show a distorted octahedral geometry around the central cobalt atom with the relevant bond angles in the range 82.5 - 95.8°, deviating from the expected 90° (**Table 4.6**). This distortion can be ascribed to the restrictions imposed by the bidentate picolinamide and β-diketonate ligand. The bond lengths are similar to those observed with the previous cobalt picolinamide β-diketonate complexes (**4.1** - **4.9**). The cobalt-pyridyl nitrogen bond lengths are in the range 1.90 - 1.93 Å, the cobalt-amide nitrogen bond lengths are in the range 1.90 - 1.94 Å and the cobalt-oxygen bond lengths in the range 1.87 - 1.92 Å. The Co-O$_{diketonate}$ bond lengths are consistent with literature values for complexes containing ferrocenyl β-diketonate ligands, with values in the range 1.84 - 1.91 Å.[48-50] There is evidence from the crystal structure for long range intermolecular hydrogen bonds, with bond distances in the range 3.06 - 3.23 Å. These hydrogen bonds tend to be either between the amide oxygen and a neighbouring pyridyl or aryl CH group or between the trifluoromethyl group and a neighbouring aryl CH group (**Figure 4.13**).
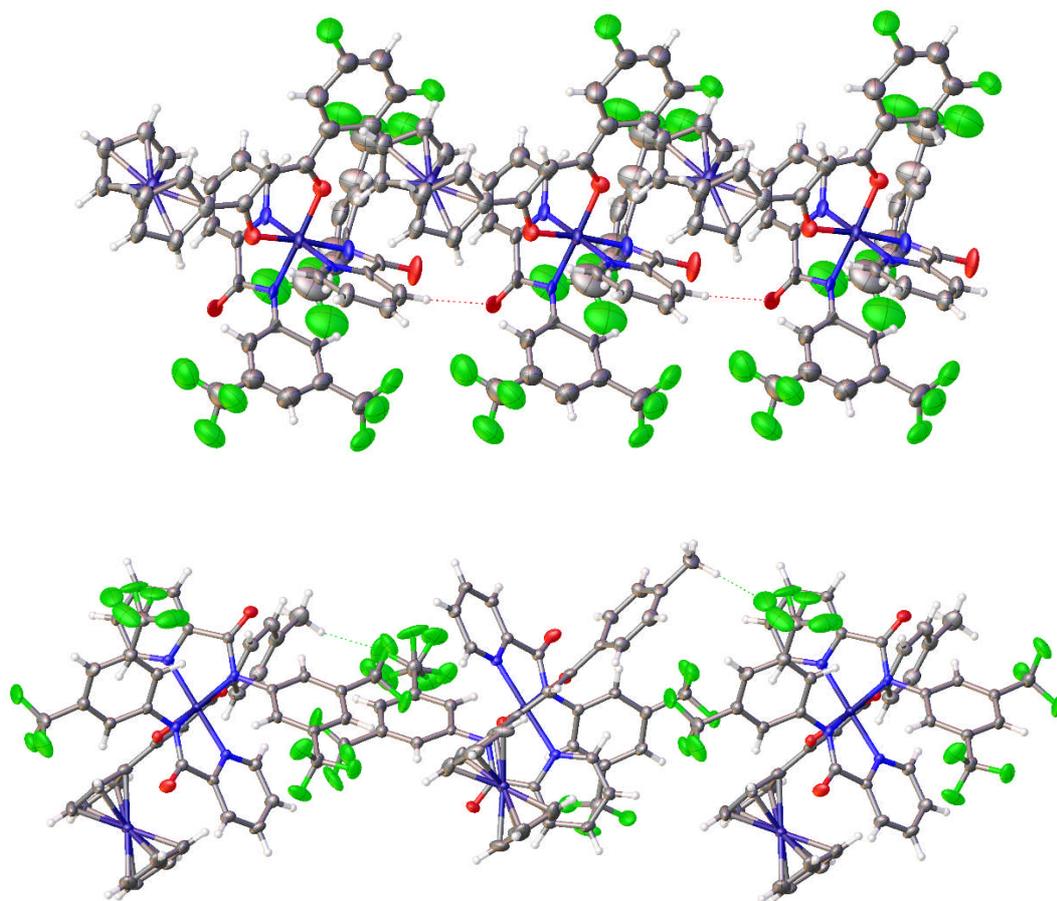
Figure 4.13: Intermolecular hydrogen bonding interactions between complexes

4.4 Conclusions

A series of novel cobalt(III) picolinamide β-diketonate complexes have been synthesised, with two picolinamide ligands (with *N,N* coordination) and a β-diketonate ligand. Within this series, two types of β-diketonate ligand have been utilised: aryl β-diketonate and ferrocenyl β-diketonate ligands. The synthesis is controlled stoichiometrically, however a number of side products limits the yields obtained. Complexes show a distorted octahedral structure with bond lengths in keeping with a cobalt(III) species. No intramolecular bonds are observed, however intermolecular hydrogen bonds are observed between neighbouring molecules. The efficacy of the complexes as anti-cancer, anti-bacterial and anti-fungal agents will be discussed in **Chapter 5**.

4.5 References

1.   F. Dimiza, A. N. Papadopoulos, V. Tangoulis, V. Psycharis, C. P. Raptopoulou, D. P. Kessissoglou and G. Psomas, *Dalton Transactions*, 2010, **39**, 4517-4528.

2.   F. Dimiza, A. N. Papadopoulos, V. Tangoulis, V. Psycharis, C. P. Raptopoulou, D. P. Kessissoglou and G. Psomas, *Journal of Inorganic Biochemistry*, 2012, **107**, 54-64.

3.   S. Tsiliou, L. A. Kefala, F. Perdih, I. Turel, D. P. Kessissoglou and G. Psomas, *European Journal of Medicinal Chemistry*, 2012, **48**, 132-142.

4.   P. B. Cressey, A. Eskandari, P. M. Bruno, C. X. Lu, M. T. Hemann and K. Suntharalingam, *Chembiochem*, 2016, **17**, 1713-1718.

5.   B. A. Teicher, M. J. Abrams, K. W. Rosbe and T. S. Herman, *Cancer Research*, 1990, **50**, 6971-6975.

6.   D. C. Ware, B. D. Palmer, W. R. Wilson and W. A. Denny, *Journal of Medicinal Chemistry*, 1993, **36**, 1839-1846.

7.   T. W. Failes and T. W. Hambley, *Dalton Transactions*, 2006, 1895-1901.

8.   T. W. Failes and T. W. Hambley, *Journal of Inorganic Biochemistry*, 2007, **101**, 396-403.

9.   G. O. Ahn, D. C. Ware, W. A. Denny and W. R. Wilson, *Radiation Research*, 2004, **162**, 315-325.

10.   J. Y. C. Chang, G. L. Lu, R. J. Stevenson, P. J. Brothers, G. R. Clark, K. J. Botting, D. M. Ferry, M. Tercel, W. R. Wilson, W. A. Denny and D. C. Ware, *Inorganic Chemistry*, 2013, **52**, 7688-7698.

11.   D. R. van Staveren and N. Metzler-Nolte, *Chemical Reviews*, 2004, **104**, 5931-5985.

12.   W. A. Wani, E. Jameel, U. Baig, S. Mumtazuddin and L. T. Hun, *European Journal of Medicinal Chemistry*, 2015, **101**, 534-551.

13.   A. Nguyen, A. Vessieres, E. A. Hillard, S. Top, P. Pigeon and G. Jaouen, *Chimia*, 2007, **61**, 716-724.

14.   S. Top, J. Tang, A. Vessieres, D. Carrez, C. Provot and G. Jaouen, *Chemical Communications*, 1996, 955-956.

15.   M. S. Allison, Unpublished results, 2017.

16.   J. C. Swarts, T. G. Vosloo, S. J. Cronje, W. C. Du Plessis, C. E. J. Van Rensbur, E. Kreft and J. E. Van Lier, *Anticancer Research*, 2008, **28**, 2781-2784.

17.   H. J. Gericke, A. J. Muller and J. C. Swarts, *Inorganic Chemistry*, 2012, **51**, 1552-1561.

18.   J. J. Hollick, B. T. Golding, I. R. Hardcastle, N. Martin, C. Richardson, L. J. M. Rigoreau, G. C. M. Smith and R. J. Griffin, *Bioorganic & Medicinal Chemistry Letters*, 2003, **13**, 3083-3086.

19.   E. V. Borisov, E. V. Skorodumov, V. M. Pachevskaya and P. E. Hansen, *Magnetic Resonance in Chemistry*, 2005, **43**, 992-998.

20.   X. Q. Han and R. A. Widenhoefer, *Journal of Organic Chemistry*, 2004, **69**, 1738-1740.

21.   L. Thomas, A. Gupta and V. Gupta, *Heterocyclic Communications*, 2002, **8**, 169-172.

22.   Y. C. Shi, H. M. Yang, W. B. Shen, C. G. Yan and X. Y. Hu, *Polyhedron*, 2004, **23**, 15-21.

23.   S. I. Dorovskikh, D. A. Piryazev, O. A. Plyusnina, L. N. Zelenina and N. B. Morozova, *Journal of Structural Chemistry*, 2014, **55**, 1075-1082.

24.   S. Pasko, L. G. Hubert-Pfalzgraf, A. Abrutis and J. Vaissermann, *Polyhedron*, 2004, **23**, 735-741.

25.   D. A. Souza, A. S. Florencio, J. Carneiro, S. S. Soriano, C. B. Pinheiro, M. A. Novak and M. G. F. Vaz, *Inorganica Chimica Acta*, 2008, **361**, 4024-4030.

26.   B. B. Aggarwal, A. Kumar and A. C. Bharti, *Anticancer Research*, 2003, **23**, 363-398.

27.   T. Esatbeyoglu, P. Huebbe, I. M. A. Ernst, D. Chin, A. E. Wagner and G. Rimbach, *Angewandte Chemie-International Edition*, 2012, **51**, 5308-5332.

28.   D. Perrone, F. Ardito, G. Giannatempo, M. Dioguardi, G. Troiano, L. Lo Russo, A. De Lillo, L. Laino and L. Lo Muzio, *Experimental and Therapeutic Medicine*, 2015, **10**, 1615-1623.

29.   K. I. Priyadarsini, *Current Pharmaceutical Design*, 2013, **19**, 2093-2100.

30.   S. Prasad, A. K. Tyagi and B. B. Aggarwal, *Cancer Research and Treatment*, 2014, **46**, 2-18.

31.   M. M. Yallapu, M. Jaggi and S. C. Chauhan, *Drug Discovery Today*, 2012, **17**, 71-80.

32.   S. Banerjee and A. R. Chakravarty, *Accounts of Chemical Research*, 2015, **48**, 2075-2083.

33.   A. Garai, I. Pant, S. Banerjee, B. Banik, P. Kondaiah and A. R. Chakravarty, *Inorganic Chemistry*, 2016, **55**, 6027-6035.

34.   A. Kareem, Laxmi, M. Arshad, S. A. A. Nami and N. Nishat, *Journal of Photochemistry and Photobiology B-Biology*, 2016, **160**, 163-171.

35.   B. M. Pires, L. C. Giacomin, F. A. V. Castro, A. D. Cavalcanti, M. D. Pereira, A. J. Bortoluzzi, R. B. Faria and M. Scarpellini, *Journal of Inorganic Biochemistry*, 2016, **157**, 104-113.

36.   A. K. Renfrew, N. S. Bryce and T. Hambley, *Chem.-Eur. J.*, 2015, **21**, 15224-15234.

37.   A. K. Renfrew, N. S. Bryce and T. W. Hambley, *Chemical Science*, 2013, **4**, 3731-3739.

38.   T. Sarkar, S. Banerjee and A. Hussain, *RSC Advances*, 2015, **5**, 16641-16653.

39.   L. J. Boucher, *Inorganic Chemistry*, 1967, **6**, 2162-2168.

40.   J. K. Clegg, D. J. Bray, K. Gloe, K. Gloe, M. J. Hayter, K. A. Jolliffe, G. A. Lawrance, G. V. Meehan, J. C. McMurtrie, L. F. Lindoy and M. Wenzel, *Dalton Transactions*, 2007, 1719-1730.

41.   A. Ali, D. Bansal and R. Gupta, *Journal of Chemical Sciences*, 2014, **126**, 1535-1546.

42.   A. Mishra, A. Ali, S. Upreti and R. Gupta, *Inorganic Chemistry*, 2008, **47**, 154-161.

43.   J. Y. Qi, H. X. Ma, X. J. Li, Z. Y. Zhou, M. C. K. Choi, A. S. C. Chan and Q. Y. Yang, *Chemical Communications*, 2003, 1294-1295.

44.   M. Ray, D. Ghosh, Z. Shirin and R. Mukherjee, *Inorganic Chemistry*, 1997, **36**, 3568-3572.

45.   M. Dakovic, D. Vila-Vicosa, M. J. Calhorda and Z. Popovic, *CrystEngComm*, 2011, **13**, 5863-5871.

46.   G. V. Tsintsadze, R. A. Kiguradze, A. N. Shnulin and K. S. Mamedov, *Journal of Structural Chemistry*, 1986, **27**, 272-278.

47.   J. Xue, X. Hua, L. Yang, W. Li, Y. Xu, G. Zhao, G. Zhang, L. Liu, K. Liu, J. e. Chen and J. Wu, *Journal of Molecular Structure*, 2014, **1059**, 108-117.

48.   D. C. Ware, P. J. Brothers, G. R. Clark, W. A. Denny, B. D. Palmer and W. R. Wilson, *Journal of the Chemical Society-Dalton Transactions*, 2000, 925-932.

49.   D. C. Ware, W. A. Denny and G. R. Clark, *Acta Crystallographica Section C-Crystal Structure Communications*, 1997, **53**, 1058-1059.

50.   D. C. Ware, W. R. Wilson, W. A. Denny and C. E. F. Rickard, *Journal of the Chemical Society-Chemical Communications*, 1991, 1171-1173.

51.   H. Zhao, X. Zhu, Y. Shang, S. Chen, B. Li and Z. Bian, *RSC Advances*, 2016, **6**, 34476-34483.

Chapter 5

Biological investigation of cobalt picolinamide complexes

5. Biological investigation

Drug discovery is the process by which new potential drugs are identified. Large chemical libraries of small molecules, natural products etc., are screened against a known biological target to identify lead molecules with the desired therapeutic effect. Further development of lead compounds is then required to optimise effects such as potency, selectivity and stability. Following lead optimisation, these drugs may progress to preclinical trials which investigate aspects such as the pharmacokinetics, pharmacodynamics and toxicity. A successful preclinical study may lead to the drug entering clinical trials. The majority of drugs which fail in clinical trials are either ineffective (or no more effective than current treatments) or are unsafe. Therefore the non-clinical phases of drug discovery and lead validation are crucial. A number of techniques are used in the screening and validation process including high throughput screening of compounds, secondary *in vitro* investigation into mechanisms of action and selectivity followed by *in vivo* analysis examining efficacy and toxicity.[1-4]

5.1 Cytotoxicity

The first stage of anti-cancer drug discovery is *in vitro* cytotoxicity screening. Cell based assays are used to measure cytotoxicity and are commonly used to screen compounds to determine their effect on target cells and thus identify any lead compounds.[3] Cytotoxicity assays determine the IC_{50} value of a compound, which is defined as the concentration of drug required to inhibit 50 % of cell proliferation. A variety of methods are used to determine the number of viable eukaryotic cells, one being tetrazolium reduction assays which are widely used due to their utility in high throughput screening.[5]

The MTT assay is a colorimetric tetrazolium reduction assay, developed by Mossman,[6] and was the first cell viability assay suitable for high throughput screening and is widely used. Viable, metabolically active cells reduce yellow MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) to purple formazan which is monitored spectrophotometrically (**Figure 5.1**).[5] Reduction of tetrazolium dyes such as MTT is dependent on NAD(P)H dependent reductases within the cell, with the concurrent oxidation of NAD(P)H to $NAD(P)^+$. The specific reductase enzymes were originally thought to be specific to the mitochondria, but now these enzymes are thought to be mainly cytosolic. Formazan accumulates within the cell as in insoluble precipitate and is deposited near the cell surface.[5, 7]
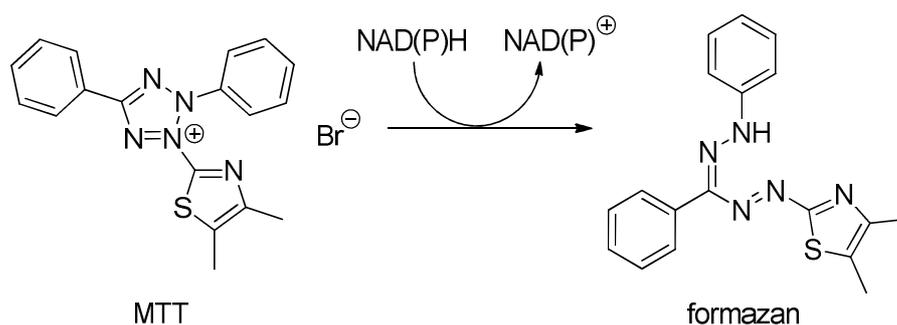
Figure 5.1: Reduction of MTT to formazan

The three series of cobalt picolinamide complexes described in **Chapter 2**, **Chapter 3** and **Chapter 4** were screened for cytotoxicity against two cancerous cell lines and a non-cancerous cell line: MIA PaCa-2 (human pancreatic carcinoma), BE (human colon carcinoma) and ARPE-19 (human retinal pigment epithelial cells) by the author and Samantha Shepherd under the supervision of Professor Roger Phillips at The University of Huddersfield. Initially, complexes were screened against all three cell lines, but as similar trends were noticed across the different cell lines it was decided to screen the remainder of the complexes against Mia PaCa-2 cells only. The complexes, at a variety of concentrations, were incubated with the cell lines at 37 °C for five days. All cytotoxicity assays were performed in triplicate. Inhibition of cell survival was determined by measuring absorbance at 540 nm following the addition of MTT and incubation at 37 °C for a further three hours. IC_{50} values are determined from graphs of percentage cell survival versus drug concentration.

Cytotoxicity screening results for cobalt(III) *tris*-picolinamide complexes, cobalt(II) *bis*-picolinamide complexes and cobalt(III) mixed ligand complexes are summarised in **Table 5.1**, **Table 5.2** and **Table 5.3** respectively. Active complexes are highlighted in green (IC_{50} < 20 µM) and partially active complexes are highlighted in orange (IC_{50} = 20 - 50 µM). Results for the active and partially active complexes only are summarised in **Figure 5.2**. Complexes **4.1**, **4.18** and **4.19** were insufficiently soluble in DMSO and cell culture medium used for the MTT assay and therefore no results could be obtained.

Table 5.1: IC$_{50}$ values of cobalt(III) *tris*-picolinamide complexes

Complex	IC$_{50}$ / μM		
	MIA PaCa-2	BE	ARPE-19
cisplatin	2.84 ± 2.05	0.66 ± 0.33	6.41 ± 0.95
2.1a	> 100	> 100	> 100
2.2	> 100	> 100	> 100
2.3	> 100	> 100	> 100
2.4	> 100	> 100	> 100
2.5	> 100	> 100	> 100
2.6	> 97.5	> 100	> 100
2.7	> 86.4	> 100	> 65.5
2.8	> 100	> 93.2	> 100
2.9	> 68.5	> 100	> 100
2.10	37.4 ± 18.9	> 74.9	> 65.4
2.11	> 62.3	> 100	> 100
2.12	> 100	> 100	> 100
2.13	17.53 ± 13.33	3.77 ± 1.98	7.57 ± 3.92
2.14a	> 100	> 100	> 100
2.15a	> 100	> 100	> 100
2.16a	> 98.1	> 100	> 100
2.17a	> 100	-	-
2.18a	> 100	-	-
2.1b	> 67.0	> 100	> 82.4
2.14b	36.73 ± 13.97	52.61 ± 6.99	29.12 ± 14.25
2.15b	55.82 ± 1.63	37.45 ± 13.04	38.67 ± 19.86
2.16b	7.38 ± 2.17	19.31 ± 1.38	6.53 ± 4.27
2.17b	> 100	-	-
2.18b	> 100	-	-

Table 5.2: IC_{50} values of cobalt(II) *bis*-picolinamide complexes

Complex	IC_{50} / µM		
	MIA PaCa-2	**BE**	**ARPE-19**
3.1	> 100	-	-
3.2	> 100	-	-
3.3	> 100	-	-
3.4	> 100	-	-
3.5	> 100	-	-
3.6	> 100	-	-
3.7	> 100	-	-
3.8	> 100	-	-
3.9	> 100	-	-
3.10	> 100	-	-
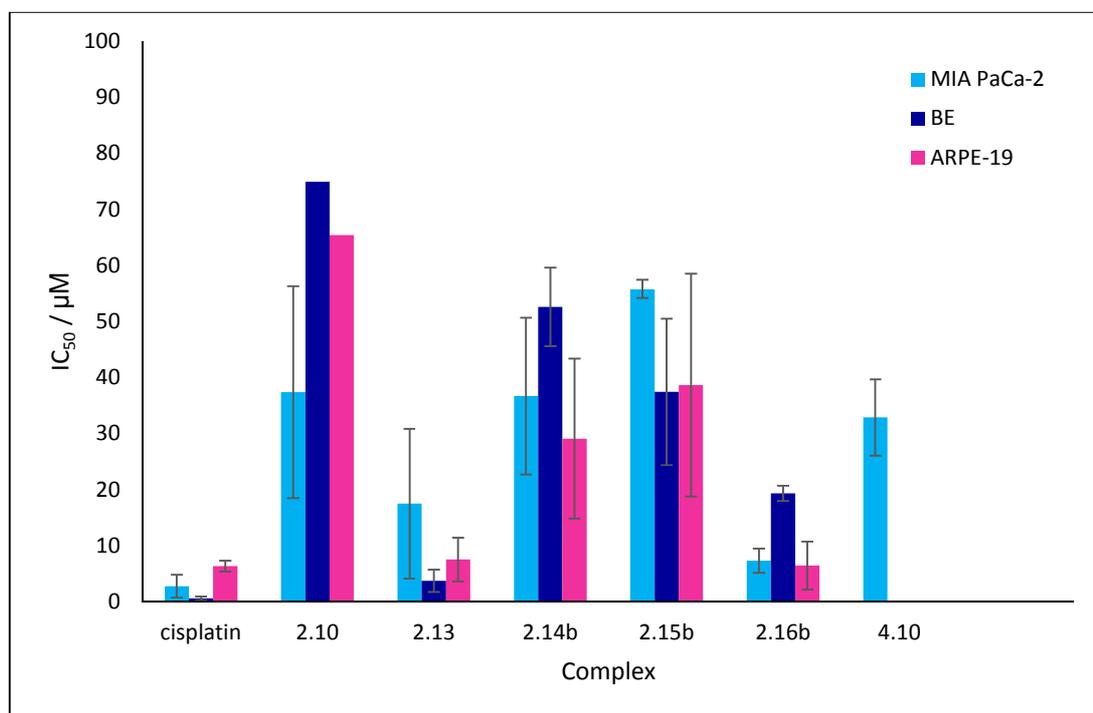
Figure 5.2: IC_{50} values of active and partially active cobalt picolinamide complexes

Table 5.3: IC$_{50}$ values of cobalt(III) mixed ligand complexes

Complex	IC$_{50}$ / µM		
	MIA PaCa-2	BE	ARPE-19
4.2	> 100	-	-
4.3	> 100	-	-
4.4	> 100	-	-
4.5	> 100	-	-
4.6	> 100	-	-
4.7	> 100	-	-
4.8	> 100	-	-
4.9	> 100	-	-
4.10	32.9 ± 6.8	-	-
4.11	> 100		
4.12	> 100	-	-
4.13	> 100	-	-
4.14	> 100	-	-
4.15	> 100	-	-
4.16	> 100	-	-
4.17	> 100	-	-

A number of complexes have cytotoxic effects against the selected cell lines, however the majority of complexes display no cytotoxic effects. This in itself is an interesting observation as cobalt ions and especially cobalt(II) chloride are known to be cytotoxic through induction of DNA damage and oxidative stress leading to apoptosis.[8-11] Three series of cobalt picolinamide complexes have been screened; five cobalt(III) *tris*-picolinamide complexes, no cobalt(II) *bis*-picolinamide complexes and one cobalt(III) mixed ligand complex show activity or partial activity. All of these complexes, with one exception, display activity against all three of the different cell lines; the implication being that complexes which are active have broad spectrum cytotoxicity against a range of different cell types and as such are not selective for cancerous cells over non-cancerous cells.

Within the series of cobalt *tris*-picolinamide complexes (**2.1a** - **2.18b**), complexes **2.13** and **2.16b** were active and complexes **2.10**, **2.14b** and **2.15b** were partially active, with potencies in the micromolar range. The general trend is that the complexes with *N,O* coordination are more active than the analogous complexes with *N,N* coordination (**Figure 5.3**). For example, complex **2.16b** with *N,O* coordination is active against all three cell lines (IC$_{50}$ = 7.38 µM against MIA PaCa-2 cells) but its *N,N* isomer complex **2.16a** is completely inactive (IC$_{50}$ > 98 µM against MIA PaCa-2 cells). The cobalt *tris*-picolinamide complexes represent true coordination isomers with no change in molecular formula, molecular weight, charge or metal oxidation state and therefore the differences in cytotoxicity derive from the differences in ligand coordination mode. This helps provide insight into the underlying mechanisms of activity for these type of complexes as previously any metal picolinamide complexes with *N,O* coordination have been associated with changes in molecular formula, molecular weight and charge due to the different ways in which the amide oxygen can coordinate to the metal centre (either in a neutral or anionic manner). The reason for this stark difference in potency between coordination isomers is unknown, possible reasons include differences in stability, hydrolysis rates, target binding etc.

This work represents the first example of metal picolinamide complexes where *N,O* coordination is associated with improved activity compared to *N,N* coordination. In fact, this is in contrast to the trend noted within a series ruthenium and osmium *p*-cymene picolinamide complexes.[12, 13] In these complexes *N,N* coordination was associated with potent activity, rapid hydrolysis and binding to guanine residues, whereas *N,O* coordination was associated with a lack of toxicity, slow hydrolysis and no binding to guanine residues.[12, 13] However, in these series of complexes the amide oxygen acts as a neutral donor resulting in a monocationic complex, this differs from the cobalt *tris*-picolinamide series in this work where amide oxygen acts as an anionic donor resulting in a neutral complex. It is therefore unclear whether the effects described previously are due to the change in ligand coordination mode, the change in charge of the complex or a combination of both.
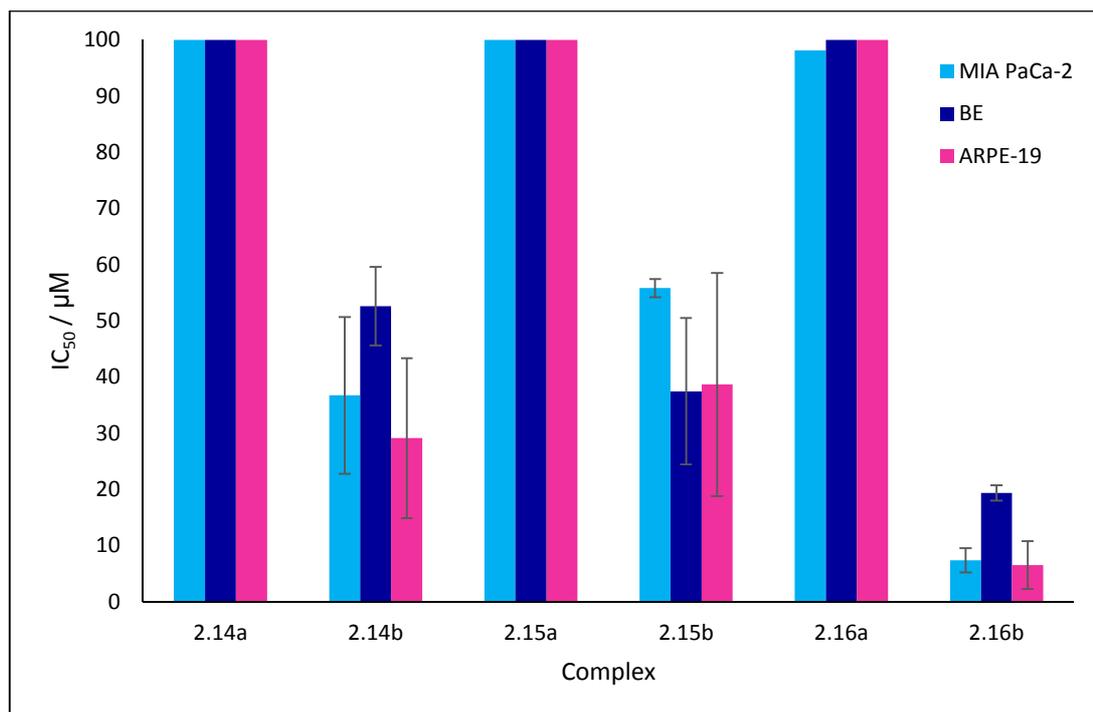
Figure 5.3: IC$_{50}$ values of **2.14b**, **2.15b** and **2.16b** with their analogous inactive isomers **2.14a**, **2.15a** and **2.16a**

The two most active complexes, designated as the lead complexes, are both doubly substituted at the *meta* position on the aryl ring with trifluoromethyl group (**2.13**) or a methyl group (**2.16b**) (**Figure 5.4**). The fact that these two similar molecules are the most active suggests that perhaps molecular shape plays an important role, where these two molecules are able to more specifically bind a target molecule; however, this would not explain why complex **2.16a**, which also has a similar shape, is inactive.

2.13   **2.16b**
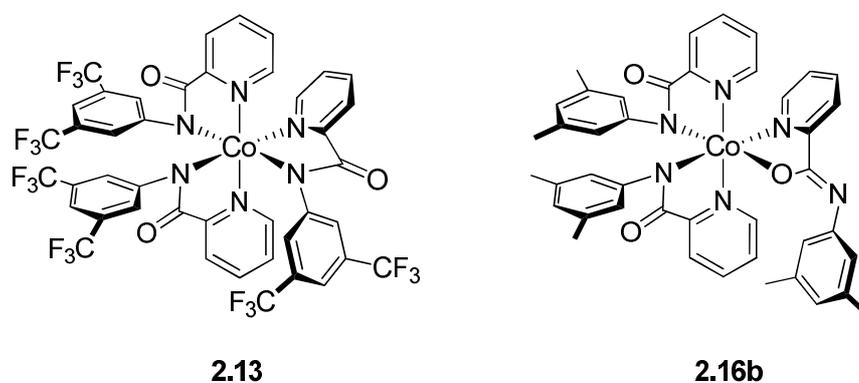
Figure 5.4: Two most active (lead) complexes

In contrast to the cobalt(III) *tris*-picolinamide complexes, all of the cobalt(II) *bis*-picolinamide complexes (**3.1** - **3.10**) were inactive. This implies that either the cobalt oxidation state or presence of three picolinamide ligands is important in relation to activity. It is thought that the shape of the molecule may be important for activity as the majority of intracellular targets are proteins which have specific structures to provides specificity to molecular interactions and reactions.[14, 15] Therefore the presence of only two picolinamide ligands may explain the non-toxic nature of the cobalt *bis*-picolinamide complexes as they may not be able to bind to the specific target due to the differences in shape.

The cobalt(III) mixed ligand complexes contain two 3,5-di(trifluoromethyl) picolinamide ligands (**1.14**) with the third ligand a β-diketonate or ferrocenyl β-diketonate ligand and are thus analogous to lead complex **2.13**. All of these complexes were inactive except for complex **4.10,** which was partially active but was less active than complex **2.13**. This implies that having the three picolinamide ligands around the central cobalt(III) ion is important for activity as substitution of just one 3,5-di(trifluoromethyl) picolinamide ligand for a β-diketonate ligand led to inactivation of the complex. This supports the suggestion that the shape of the molecule is important for activity. The partially active complex **4.10** contains a CHF_2 group as the ferrocenyl β-diketonate substituent. Complexes **4.11** and **4.12,** with a trifluoromethyl or methyl group respectively in the same position as the CHF_2 group in complex **4.10,** are inactive which suggests that for this molecule electronics rather than sterics plays a role in activity. The lack of activity for complex **4.11** is surprising as the ferrocenyl β-diketonate ligand which constitutes part of this complex is cytotoxic as the free ligand.[16] Other than this example, there is no improvement in activity with addition of the ferrocene group which is different to several examples in organometallic chemistry where addition of ferrocene improves drug activity.[17, 18]

Cobalt picolinamide complexes are quite sparse within the literature and only a few have been tested for their anti-cancer potential. Only a series of cobalt(III) heterobimetallic complexes (with cadmium, mercury or zinc) containing tridentate picolinamide ligands have been screened against cancerous U87 (human glioblastoma) and non-cancerous HEK (human embryonic kidney) cell lines.[19] These complexes displayed moderate activity, in the micromolar or millimolar range, against both the cancerous and non-cancerous cell lines. These cytotoxicities are less potent than for the cobalt picolinamide complexes investigated in this chapter, however the trend of broad spectrum toxicity rather than cancer cell selectivity is consistent with that observed for these cobalt picolinamide complexes.

5.2 Hypoxia studies

The ability to preserve oxygen (O_2) homeostasis is essential for cell survival. Hypoxia defines an environment of reduced oxygen concentration. Mammalian cells typically exist at 2 - 9 % oxygen (normoxia) with hypoxia defined as ≤ 2 % oxygen and severe hypoxia (anoxia) as ≤ 0.02 % oxygen.[20] Hypoxia is present in many diverse pathologies such as infarction, ischaemia, stroke and cancer.

Tumours which are larger than 1 mm^3 contain hypoxic regions due to insufficient vascularisation of the rapidly growing tumour. Cancer cells are able to survive their hypoxic state through activation of hypoxia-inducible factor-1 (HIF-1). HIF-1 is a heterodimeric transcription factor consisting of an α and β-subunit. In normoxia, the α-subunits are hydroxylated, allowing ubiquitination and degradation by the proteasome. In hypoxia, the specific hydroxylase enzyme is inhibited and thus the α-subunits are not degraded.[20-24] The intact HIF-1 can bind to the hypoxia responsive element (HRE) and upregulate transcription of a number of genes involved in cell survival such as genes encoding for proteins involved in glycolysis, angiogenesis and the inflammatory response.[23, 25, 26] HIF-1 is implicated in a number of cancers and there is also a strong correlation between HIF-1 overexpression and the tumour grade and vascularisation.[22, 27]

Tumour hypoxia is a strong prognostic factor in many cancers; extensive hypoxia results in reduced chemotherapeutic drug delivery, intrinsic resistance to radiotherapy, neovascularisation, greater metastatic capacity and increased apoptotic resistance.[20, 22, 28] Therapies which destroy normoxic but not hypoxic cancer cells are more likely to result in recurrence. The hypoxic environment of these cells may also lead to upregulation of genes involved in drug resistance and promotion of cell growth.[29, 30] Hypoxic cancer cells also present a more reducing environment, thought to help stabilise HIF-α, compared to normoxic non-cancerous cells, providing the potential for selective targeting of hypoxic cancer cells.

A novel approach to overcome the resistance of hypoxic cells to cancer therapies is to employ bio-reductive prodrugs. Metal complexes are well suited to such strategies due to the variations in coordination number, geometry and accessible oxidation states.[31] Cobalt(III) prodrugs have already been discussed in **Chapter 1**. These tend to consist of a central cobalt(III) ion with one known cytotoxic ligand which is released upon reduction to cobalt(II). Other transition metal prodrugs under

investigation include platinum(IV),[32] ruthenium(III),[33, 34] copper(II)[35] and iron(III)[36] prodrugs.

Cobalt(III) *tris*-picolinamide complexes were screened for cytotoxicity under hypoxic conditions against MIA PaCa-2 cells by the author and Samantha Shepherd under the supervision of Professor Roger Phillips at The University of Huddersfield. The MTT assay was performed as previously described (**Chapter 5.1**) except all cell culturing and cell incubation was performed in an environment of 0.1 % oxygen. The cytotoxicity results in normoxia (21 % oxygen) and hypoxia are summarised in **Table 5.4**. Results for complexes which are active or partially active are also displayed in **Figure 5.5**.
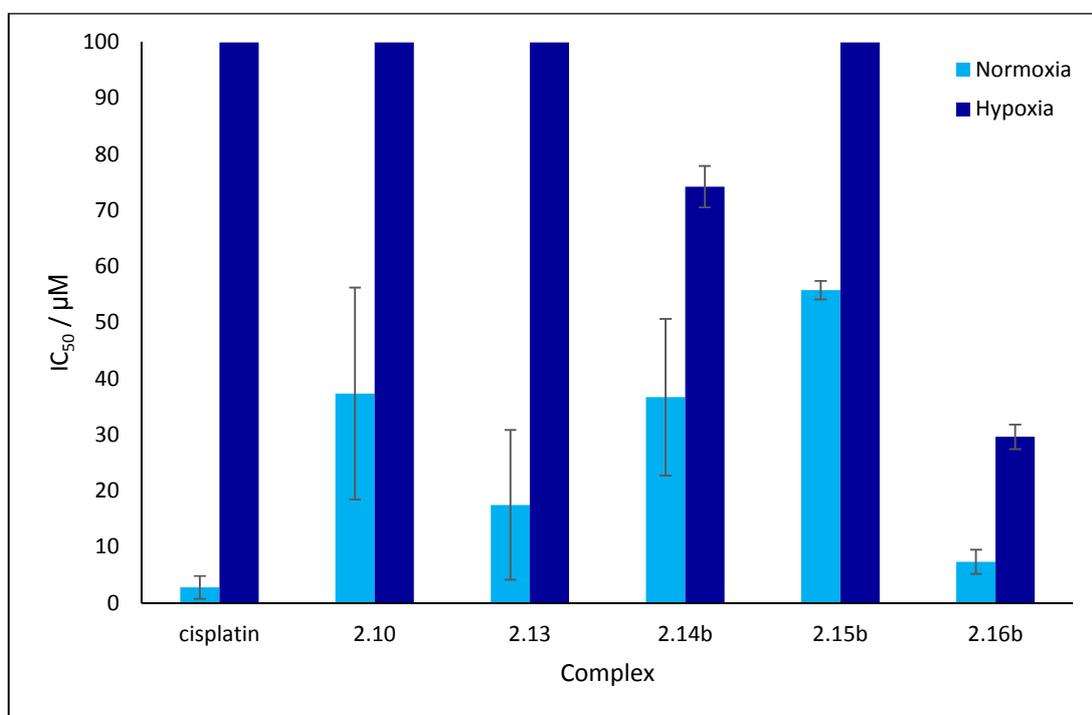
Figure 5.5: IC$_{50}$ values of active and partially active complexes under normoxic and hypoxic conditions

Table 5.4: IC_{50} values of cobalt(III) *tris*-picolinamide complexes under normoxic and hypoxic conditions

Complex	IC_{50} / µM	
	Normoxia	**Hypoxia**
cisplatin	2.84 ± 2.05	> 100
2.1a	> 100	> 100
2.2	> 100	> 100
2.3	> 100	> 100
2.4	> 100	> 100
2.5	> 100	> 78.3
2.6	> 97.5	> 100
2.7	> 86.4	> 100
2.8	> 100	> 100
2.9	> 68.5	> 95.3
2.10	37.4 ± 18.9	> 100
2.11	> 62.3	> 100
2.12	> 100	> 100
2.13	17.53 ± 13.33	> 100
2.14a	> 100	> 100
2.15a	> 100	> 100
2.16a	> 98.1	> 100
2.1b	> 67.0	> 100
2.14b	36.73 ± 13.97	74.25 ± 3.68
2.15b	55.82 ± 1.63	> 100
2.16b	7.38 ± 2.17	29.68 ± 2.18

All complexes were less toxic to MIA PaCa-2 cells under hypoxic conditions compared to normoxic conditions. Three of the five active or partially active complexes, **2.10**, **2.13** and **2.15b**, became completely inactive ($IC_{50} > 100$ µM) in hypoxia. Complex **2.16b** remained at least partially active ($IC_{50} = 29.68$ µM) even under the low oxygen conditions. Cells under hypoxic conditions have a more reducing intracellular

environment than when under normoxic conditions and thus there is the potential for the intracellular bioreduction of cobalt(III) to cobalt(II) with the concurrent release of one ligand. If the cobalt(III) *tris*-picolinamide complexes are being reduced to cobalt(II) *bis*-picolinamide complexes, this explains the decrease in potency. All of the synthesised cobalt(II) *bis*-picolinamide complexes were non-toxic, therefore reduction of the cobalt(III) complexes to similar species would be accompanied by a reduction in the cytotoxicity. A number of cobalt(III) prodrugs utilise this reduction to target cancer cells, however these all tend to contain a known cytotoxic ligand which is released upon reduction to cobalt(II).[37-39] Interestingly, complex **2.16b** retains activity even in hypoxic conditions despite the fact that it does not contain a ligand which is known to be cytotoxic. This perhaps implies that this compound is not fully intracellularly reduced from cobalt(III) to cobalt(II), due to a reduction potential outside of the biological range, thus retaining some of the active complex.

5.3 Cell viability and cell cycle analysis

Once a lead compound has been identified, the next step in the drug development process involves elucidating the mechanism of action.[3, 4] Anti-cancer drugs may act through induction of cancer cell death (cytotoxic drugs) e.g. cisplatin, or through inhibition of cancer cell proliferation (cytostatic drugs) e.g. tamoxifen. Biological assays coupled to cell imaging techniques, such as flow cytometry, can discriminate between these two different types of effect. Following determination of cytotoxic or cytostatic action, further investigations include apoptosis assays or cell cycle analysis.[5, 40, 41]

Cytostatic drugs inhibit the cell cycle to halt cell proliferation. The cell cycle is the sequence of events that occur during the lifetime of a cell leading to cell division. In eukaryotic cells, the cell cycle is divided into four phases: mitosis (M phase), gap-1 (G_1 phase), synthesis (S phase) and gap-2 (G_2 phase) (**Figure 5.6**). Normal non-dividing cells are quiescent (G_0 phase); when cells enter the cell cycle they enter in G_1 phase, the growth phase when the cell increases in size, followed by S phase when DNA is replicated. This is followed by G_2 phase, a further short period of cell growth, and finally M phase when mitosis occurs, separating the chromosomes and dividing the cell to produce two daughter cells. The cell cycle is very tightly regulated by a number of different pathways which controls progression through the cell cycle, these are frequently disrupted during the oncogenic transformation of a cell.[42-46]

139
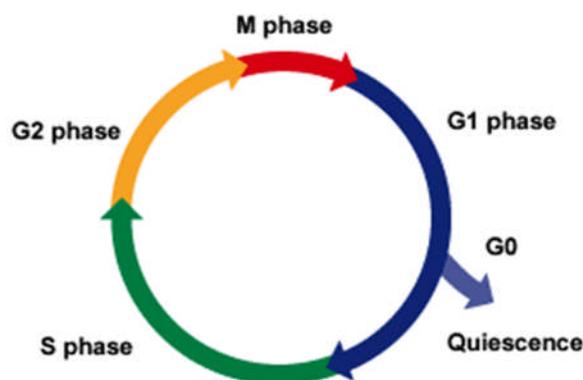
Figure 5.6: The eukaryotic cell cycle[47]

Different drugs inhibit different phases of the cell cycle but tend to operate during S phase, M phase or all phases. Alkylating agents, e.g. cisplatin and other platinum drugs, nitrogen mustards and nitrosoureas, directly damage DNA and operate at all phases of the cell cycle. Cytotoxic antibiotics, e.g doxorubicin, mitomycin-C and bleomycin, also directly alter DNA and operate at all phases of the cell cycle. Anti-metabolites, e.g. 5-fluorouracil, 6-mercaptopurine and methotrexate, substitute for normal metabolites to interfere with DNA replication and thus operate at S phase. Topoisomerase inhibitors, e.g. topotecan, etoposide and mitoxantrone, inhibit enzymes involved in DNA unwinding for replication and thus operate at S phase. Mitotic inhibitors, e.g. taxanes, vincristine and vinblastine, prevent formation of the mitotic spindle often through binding to tubulin subunits and therefore act during M phase.[44, 45, 48, 49]

The two lead complexes (**2.13** and **2.16b**) were tested for their effects on cell viability and the cell cycle of MIA PaCa-2 cells by the author and Samantha Shepherd under the supervision of Professor Roger Phillips at The University of Huddersfield. The complexes, at a variety of concentrations, were incubated with MIA PaCa-2 cells at 37 °C for five days. For cell viability assay, single cell suspensions were prepared and stained with acridine orange and 4',6-diamidino-2-phenylindole (DAPI) to enable counting of the entire cell population and non-viable cells by an automated cell counter. For cell cycle analysis, single cell suspensions were washed with phosphate-buffered saline (PBS), treated with lysis buffer and DAPI and cellular fluorescence quantified using an image cytometer. Markers of cellular content and DNA content are used to demarcate cells in different stages of the cell cycle.[50]

The cell viability results for complexes **2.13** and **2.16b** are shown in **Table 5.5** and **Table 5.6** and in **Figure 5.7**. The cell cycle analyses for complexes **2.13** and **2.16b** are shown in **Table 5.7** and **Figure 5.8**.

Table 5.5: Cell viability summary for complex **2.13**

	Concentration / µM			
	0	**5**	**10**	**20**
Total cells	3.7×10^6	3.9×10^6	3.6×10^6	2.54×10^6
Cell viability / %	81.8	79.5	85.4	85
Cell diameter / µm	18.3 ± 11.3	17.5 ± 10.8	16.7 ± 10.7	17.8 ± 11.9
Aggregates of > 5 cells	65	61	59	54

Table 5.6: Cell viability summary for complex **2.16b**

	Concentration / µM			
	0	**5**	**10**	**20**
Total cells	3.7×10^6	3.82×10^6	3.6×10^6	2.63×10^6
Cell viability / %	81.8	76	76	86
Cell diameter / µm	18.3 ± 11.3	16.5 ± 10.1	17.2 ± 10.8	17.6 ± 10.8
Aggregates of > 5 cells	65	57	51	54

For both complexes there is a reduction in the number of total cells but only at concentrations above the respective IC_{50} values (20 µM) and there is no reduction in the percentage of viable cells. There are also no readily observable changes in cell morphology, with cell shape and structure remaining consistent throughout (**Figure 5.7**). Cytotoxic drugs cause cell death and thus decrease the number of total cells and the percentage of viable cells. Cytostatic drugs halt cell proliferation and thus do not change the total number of cells (over a short time frame) or the percentage of viable cells. These results suggest that complexes **2.13** and **2.16b** act as cytostatic agents through interruption of the cell cycle.

Complex **2.13**   Complex **2.16b**

a)

b)

c)

d)
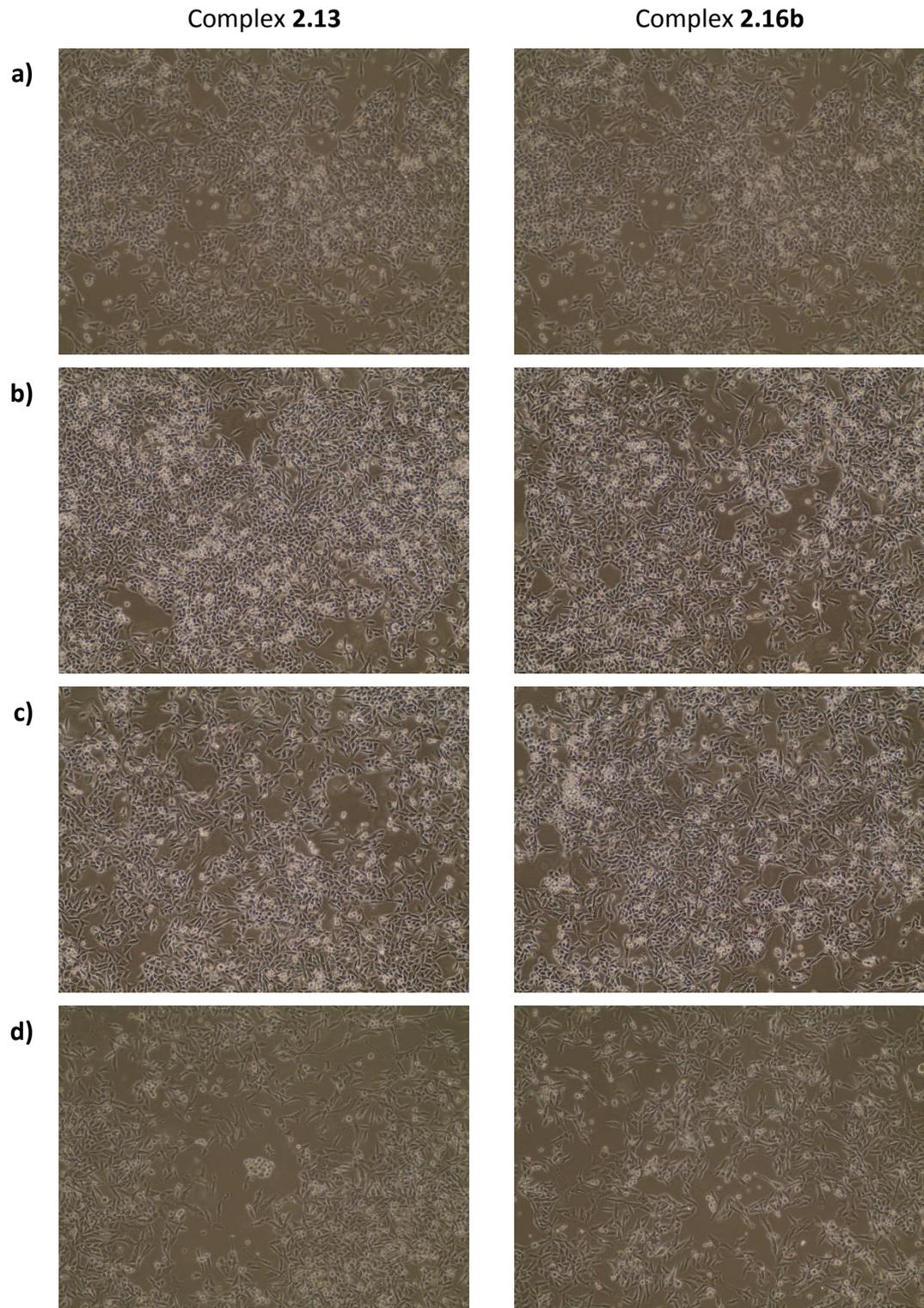
Figure 5.7: Representative optical microscopy images (× 2) of MIA PaCa-2 cells in the presence of complexes **2.13** and **2.16b** at various concentrations a) 0 µM b) 5 µM c) 10 µM d) 20 µM

Table 5.7: Cell cycle analysis for complexes **2.13** and **2.16b**

Cell cycle phase	Cell population / %							
	Concentration of complex 2.13 / µM				Concentration of complex 2.16b / µM			
	0	5	10	20	0	5	10	20
Sub G_1	10	11	10	10.5	10	9	8	11.2
G_1	83	83.4	83.3	74.2	83	84.3	85.3	76
S	4	3.6	3.4	6.3	4	3.4	3.4	6.3
M	3	2	3.3	9	3	3.3	3.3	6.5
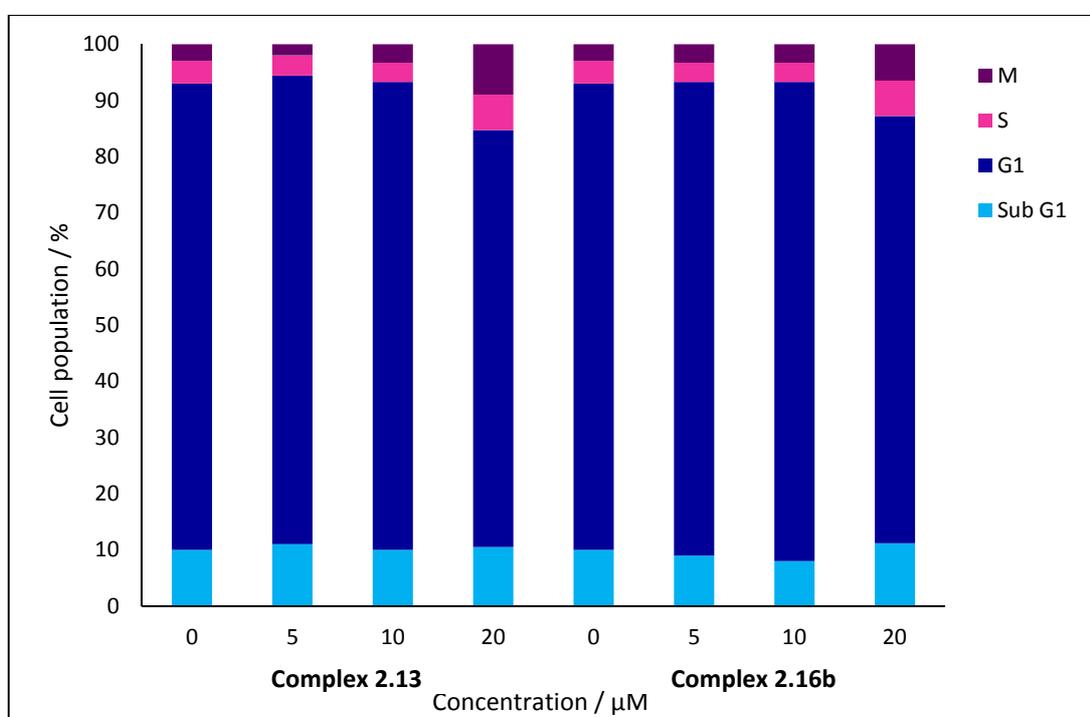
Figure 5.8: Graphical representation of cell cycle analysis for complexes **2.13** and **2.16b**

Cell cycle analysis shows that at high concentrations of both complexes (20 µM) there is a small increase in the population of cells in S phase and a larger increase in the percentage of cells in M phase. Even small changes in the percentage of cells in these phases is significant as the majority of cells are in G_1 phase. The percentage of cells in M phase has tripled (3 % to 9 %) for complex **2.13** and doubled (3 % to 6.5 %) for complex **2.16b**. This is coupled with a reduction of cells which are in G_1 phase. This demonstrates that both complexes initiate a level of cell cycle arrest during M

phase. M phase is divided into two processes: mitosis and cytokinesis. In mitosis, the replicated chromosomes condense and attach to mitotic spindle fibres which pull each set of chromosomes to different sides of the cell. This is immediately followed by cytokinesis in which a contractile ring forms and contracts inwardly to divide the cell (with its constituent chromosomes, organelles, cytoplasm and cell membrane) into two to produce two identical daughter cells.[14, 42] There are two classes of anti-cancer drugs which target M phase: taxanes and vinca alkaloids, which are both natural product derivatives. These M phase inhibitors act through disruption of mitotic spindle formation through interference with tubulin polymerisation.[44, 48] There are a few examples of inorganic complexes, mainly ruthenium arenes or ferrocenyl analogues of known inhibitors, which cause M phase arrest either through interruption of signalling pathways or inhibition of tubulin polymerisation.[51-54]

As there is only a low proportion of cells in M phase at a given time point any changes in this population of cells are inherently small. Therefore, further investigation of these complexes at the higher concentration of 50 µM was performed, following incubation for five days, in order to determine if the M phase arrest is replicated and if the effect is dose dependent. The cell viability results for complexes **2.13** and **2.16b** at a concentration of 50 µM are shown in **Table 5.8** and in **Figure 5.9**. The cell cycle analysis for complex **2.13** at a concentration of 50 µM is shown in **Table 5.9** and **Figure 5.10**.

Table 5.8: Cell viability summary for complexes **2.13** and **2.16b** at 50 µM.
* = unable to determine due to low total cell count

	Complex		
	Control	**2.13**	**2.16b**
Total cells	3.13×10^5	2.34×10^5	2.52×10^3
Cell viability / %	91.3	67.5	87.5
Cell diameter / µm	18.4 ± 9.8	18.0 ± 12.0	*
Aggregates of > 5 cells	4	7	0
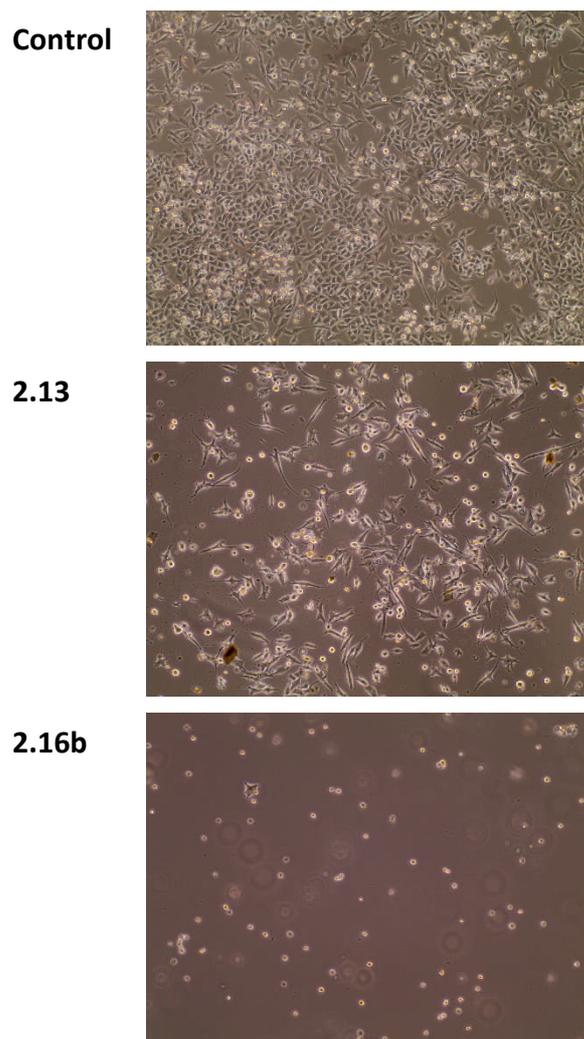
Figure 5.9: Representative optical microscopy images (× 5) of MIA PaCa-2 cells with no complex and in with complexes **2.13** and **2.16b** at 50 μM

Table 5.9: Cell cycle analysis for complex **2.13** at 50 μM

Cell cycle phase	Cell population / %	
	Concentration / μM	
	0	50
Sub G_1	9	6
G_1	64	44
S	15	24
M	12	23
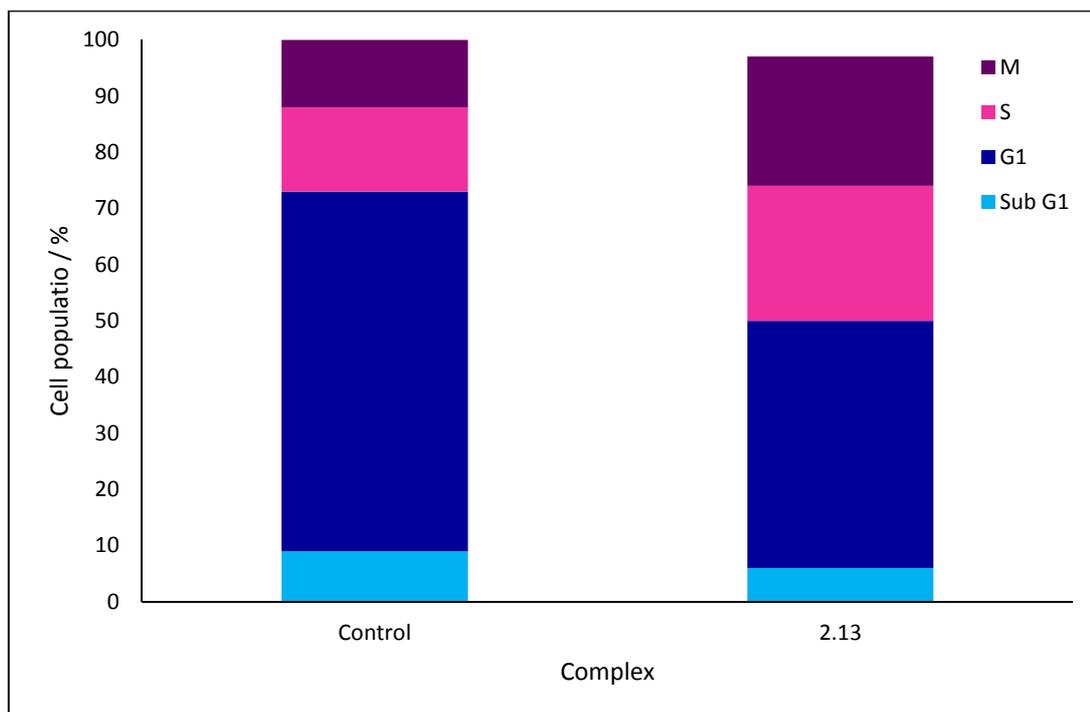
Figure 5.10: Graphical representation of cell cycle analysis for control and complex **2.13** at 50 µM

At the higher concentration of 50 µM, complex **2.13** shows a small reduction in the number total cells with a 25 % reduction in the percentage of viable cells. Complex **2.16b** shows a 100-fold reduction in the total number of cells with no reduction in the percentage of viable cells, however this may be due to the low cell count. There are also no readily observable changes in cell morphology for complex **2.13** compared to the control; however for complex **2.16b** not only is there an obvious change in cell morphology to smaller more spherical cells but the decrease in cell population is also readily observable. For complex **2.13** the results are similar to when lower concentrations were analysed, implying that this complex remains a cytostatic agent which acts through interruption of the cell cycle. However, for complex **2.16b** the results are different to when lower concentrations were analysed. At higher concentrations complex **2.16b** appears to induce cell death, i.e. acts as a cytotoxic rather than a cytostatic agent.

Cell cycle analysis could only be performed for complex **2.13** due to the low number of total cells present after incubation with complex **2.16b**. There is an increase in the population of cells in S phase and a larger increase in the percentage of cells in M phase. In fact the percentage of cells in M phase doubled (12 % to 23 %), this is coupled with a reduction of cells which are in G_1 phase. This demonstrates that even

at higher concentrations the mechanism of action of complex **2.13** appears to be the induction cell cycle arrest during M phase.

5.4 Cancer stem cell studies

Cancer stem cells (CSCs) are a sub-type of tumour initiating cancer cells with stem cell-like properties, such as the ability to self-renew and differentiate. The origination of CSCs is under debate, but the two main hypotheses state either that CSCs originate from the oncogenic transformation of normal stem cells or from partially differentiated progenitor cells.[55-58] Conventional cancer therapies are not effective against CSCs as these target more rapidly growing cell types which make up the bulk of the tumour. While tumour mass is reduced, the CSCs persist and are able to generate new, often aggressive, tumours at the primary or secondary sites.[58, 59] In order for cancer treatment to be more effective and avoid relapse, it is crucial to be able to remove CSCs in addition to the main tumour bulk. Potential drug targets for CSCs include deregulated signalling pathways, cell surface markers and dysfunctional organelles.[60-63] Investigation into CSC-specific drugs has led to number of potential drugs in clinical or pre-clinical trials such as salinomycin, the current gold standard for CSC-selective agents.[63, 64]

Research into the anti-CSC potential of metal-based drugs is still in its infancy. To date a limited number of cobalt,[65] copper,[66, 67] iron,[68] vanadium,[69] gold[70] and osmium[71] complexes have been investigated. The only cobalt complexes so far investigated are cobalt(III) cyclams, one of which contains two naproxen molecules (a nonsteroidal anti-inflammatory drug (NSAID)) as axial ligands (**Figure 5.11**). The cobalt(III) cyclam moiety acts as a prodrug which is activated by reduction to cobalt(II) in hypoxic environments, such as a cancer cell or cancer stem cell.[31] The cobalt cyclam naproxen complex showed selective activity for breast CSC-enriched cells and additionally inhibited the formation of the small tumour-like mammospheres with concurrent reduction in viability.[65]
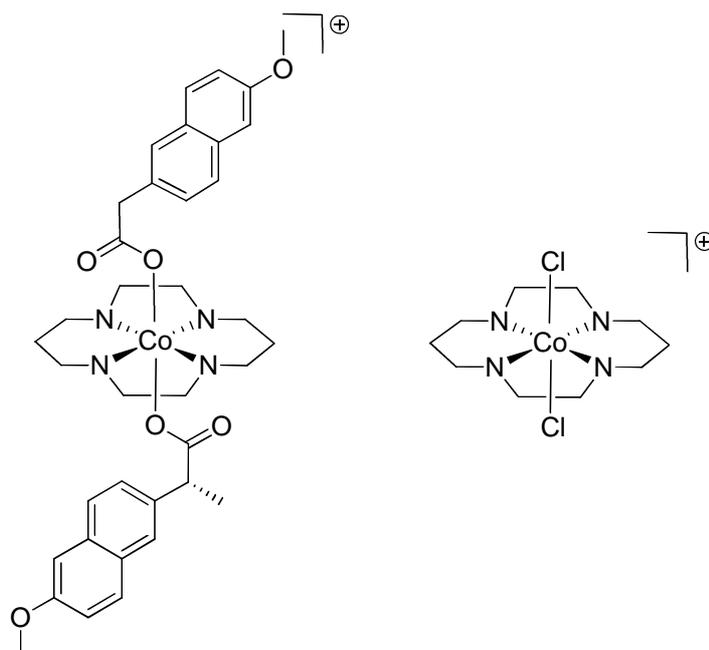
Figure 5.11: Cobalt cyclam complexes investigated against CSCs

The two lead complexes (**2.13** and **2.16b**) were tested for their anti-CSC activity by Paul Cressey under the supervision of Dr. Kogularamanan Suntharalingam at King's College London. The IC_{50} values were determined using the five day MTT assay as previously described (**Chapter 5.1**) against two cancerous human mammary epithelial cell lines: HMLER (stable CSC-like population of 5 - 8 %) and HMLER-shEcad (stable CSC-like population of approximately 90 %).[72] Additionally, IC_{50} values were determined with mammospheres, which are three-dimensional tumour-like structures which are able to form in non-adherent, serum-free media. The ability of the complexes to inhibit the formation of mammospheres was determined. Single-cell suspensions of CSC-enriched HMLER-shEcad cells were incubated with the complexes, at their IC_{20} values, for five days and mammosphere formation determined microscopically. The ability to inhibit mammosphere formation in single cell suspensions is used as a marker for anti-CSC potency.[65, 73]

The IC_{50} values of complexes **2.13** and **2.16b** are shown in **Table 5.10** and the results from the mammosphere formation assay are shown in **Figure 5.12** and **Figure 5.13**.

Table 5.10: IC$_{50}$ values of lead complexes against CSCs

Complex	IC$_{50}$ / µM		
	HMLER	HMLER-shEcad	Mammosphere
2.13	0.49 ± 0.06	0.52 ± 0.05	1.57
2.16b	1.3 ± 0.1	0.88 ± 0.02	8.39

Complexes **2.13** and **2.16b** are active against both the CSC enriched and non-CSC enriched cancer cells with micromolar or sub-micromolar potencies. Complex **2.13** has greater potency against both cell types compared to complex **2.16b**; however, complex **2.16b** is moderately selective for CSC enriched cancer cells (IC$_{50}$ = 0.88 µM) over the bulk cancer cells (IC$_{50}$ = 1.3 µM). In addition, both complexes effectively reduce the viability of mammospheres at micromolar concentrations, with complex **2.13** the more active of the two (IC$_{50}$ = 1.57 µM) (**Table 5.10**). These results are similar to the cobalt cyclam complex with naproxen (**Figure 5.11**) which also displays submicromolar IC$_{50}$ values, however this complex displays greater selectivity for CSC enriched cancer cells over bulk cancer cells than either complex **2.13** or **2.16b**.[65]
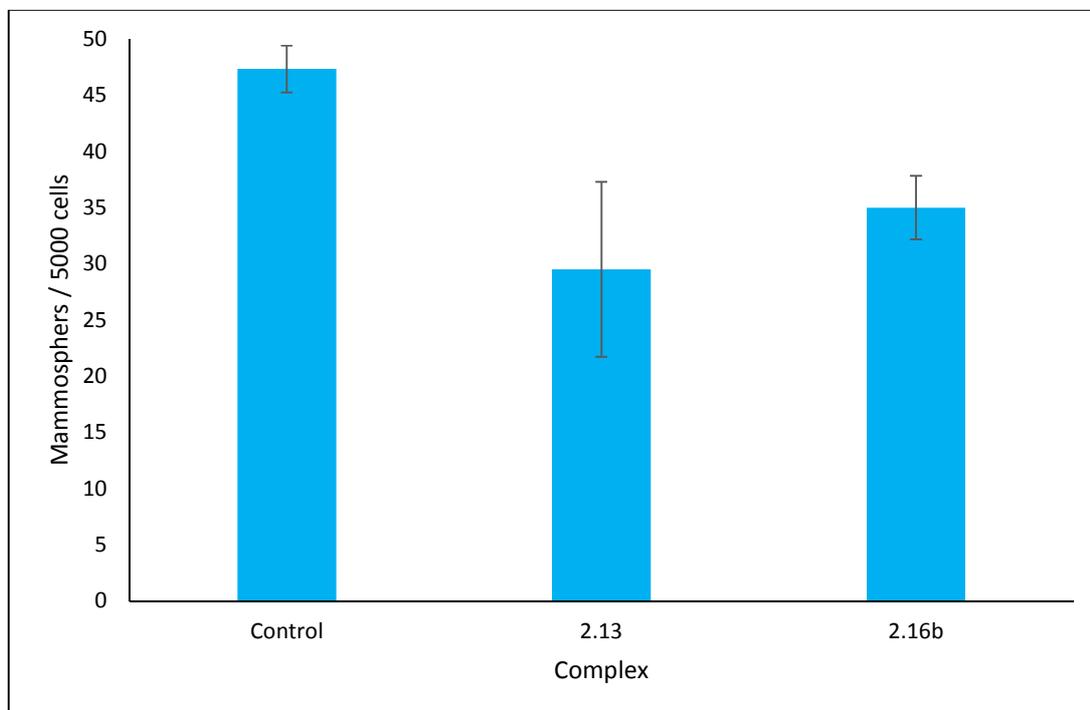
Figure 5.12: Mammosphere formation in absence or presence of complexes **2.13** and **2.16b**
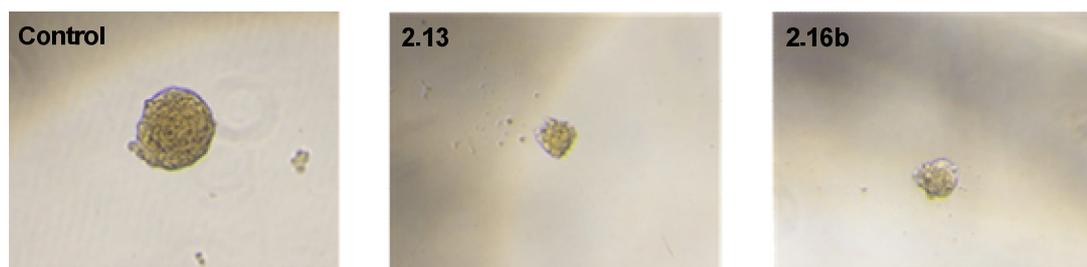
Figure 5.13: Representative bright-field microscopy images (× 10) of mammospheres in absence or presence of complexes **2.13** and **2.16b**

Both complexes reduced the number of mammospheres formed from single cell suspensions of HMLER-shEcad cells compared to untreated cells (**Figure 5.12**). Complexes **2.13** and **2.16b** both resulted in a 25 - 40 % decrease in the number of mammospheres formed. In addition, mammosphere size decreased upon incubation with both complexes (**Figure 5.13**). As the ability to inhibit mammosphere formation is a marker for CSC potency, these results confirm that both complexes are active against CSCs. The decrease in the number and size of mammospheres is again consistent with the previously studied cobalt complex, however this displayed a greater reduction in the number of mammospheres of 65 %.[65]

5.5 Anti-bacterial activity

Antibiotic resistance is one of the key challenges facing modern day medicine.[74] There is growing resistance amongst both Gram-negative and Gram-positive bacteria, including a group of pathogens which constitute the majority of hospital based infections- the ESKAPE pathogens. The ESKAPE pathogens are *Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa* and *Enterobacter* species, a number of which are resistant to multiple drugs.[75, 76] Bacteria are able to acquire resistance through various mechanisms such as drug inactivation, modification of drug binding sites, changes in cell permeability and biofilm formation.[77] Resistance can spread rapidly due to the ability of bacteria to transfer genes both vertically (between generations) and horizontally (within the same generation).[78] New antibiotics with wide ranging action against multi-drug resistant pathogens are needed; however, the number of new approved antibiotics has dramatically slowed.[79, 80] Most of these new drugs are improved versions of drugs from pre-existing antibiotic drug classes which merely delays the problem of resistance rather than offering a solution. Therefore it is

important to develop whole new classes of antibiotic with novel modes of action, this has led to the investigation of organometallic and coordination complexes as potential drugs.

A number of cobalt complexes with anti-bacterial activity have been reported in the literature, starting with the bacteriostatic and bactericidal activity of a cobalt(III) cationic coordination complex reported by Dwyer in 1950s.[81] A variety of cobalt(II) and cobalt(III) complexes with a number of diverse ligands have been investigated.[82] Cobalt(III) complexes with bidentate and tridentate picolinamide including a set of heterobimetallic complexes have been investigated by Mishra (**Figure 5.14**). The bidentate and tridentate complexes showed activity against resistant strains of *Pseudomonas* and *E. coli* and against standard strains of *Shigella* and *Klebsiella* (MIC = 5 - 350 μg ml^{-1}). It is suggested that the presence of uncoordinated pyridyl rings is responsible for the anti-bacterial activity.[83] Some of the heterobimetallic complexes were effective against *P. vulgaris*, *E. coli* and *P. aeruginosa* (MIC = 5 - 350 μg ml^{-1}). The authors suggest that the anti-bacterial activity may be due to the presence of labile sites on the second metal ion.[19]
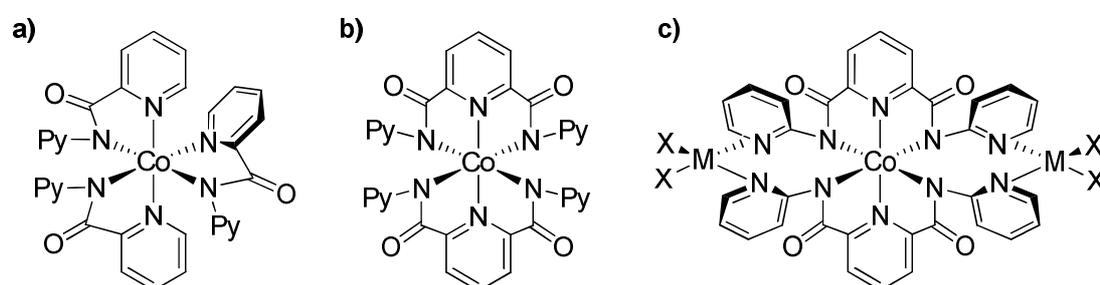
Figure 5.14: Cobalt picolinamide complexes investigated as anti-bacterial agents a) cobalt bidentate complexes b) cobalt tridentate complexes c) heterobimetallic complexes

Complexes were screened for their anti-bacterial activity against *Escherichia coli* (*E. coli*), *Klebsiella pneumoniae* (*K. pneumoniae*), *Acinetobacter baumannii* (*A. baumannii*), *Pseudomonas aeruginosa* (*P. aeruginosa*) and *Staphylococcus aureus* (*S. aureus*) by The Community for Antimicrobial Drug Discovery (CO-ADD) at The University of Queensland. The complexes, at a single concentration of 32 μg mL^{-1}, were incubated with the bacterial strains at 37 °C for 18 hours without shaking. All growth inhibition assays were performed in duplicate. Growth inhibition was determined by measuring absorbance at 600 nm. Complexes with growth inhibition

values greater than 80 % are classed as active and complexes with growth inhibition values in the range 50- 80 % are classed as partially active.

Anti-bacterial activity screening results for cobalt(III) *tris*-picolinamide complexes, cobalt(II) *bis*-picolinamide complexes and cobalt(III) mixed ligand complexes are summarised in **Table 5.11**, **Table 5.12** and **Table 5.13** respectively. Partially active complexes are highlighted in orange. Complexes **4.1**, **4.18** and **4.19** were insufficiently soluble in DMSO and cell culture medium used for the growth inhibition assays and therefore no results could be obtained.

Only one complex displayed any notable activity against any of the bacterial strains screened; all other complexes were inactive. Complex **2.8**, a cobalt(III) *tris*-picolinamide complex (*N,N* coordination) with a *para*-chloro substituent on the aryl ring, was partially active against *S. aureus* with growth inhibition of 65.9 %. Interestingly, complex **2.8** shows selective activity against *S. aureus* and is inactive against the other four bacterial strains. Alteration of the position of the chloro substitution to the *meta* position resulted in inactivation of the complex and conversion of the chloro to a fluoro or bromo also resulted in loss of activity. The *para*-chloro substituent appears to be crucial for the selective toxicity to *S. aureus*.

These results contrast with the previous bidentate cobalt picolinamide complexes studied (**Figure 5.14a**) which were all active against *E. coli*, *Klebsiella* and *Pseudomonas*.[83] The related tridentate cobalt complexes and related heterobimetallic complexes also displayed broad anti-bacterial activity.[19, 83] With regards to the bidentate and tridentate complexes, it is hypothesised that the uncoordinated pyridyl rings present in these complexes is responsible for their activity, due to the ability of these pyridyl rings to form strong interactions with cations.[83] These results tend to support this hypothesis as none of the complexes tested contained coordinating groups within or attached to the aryl ring and all besides one complex were inactive. In addition, the previously tested complexes were active against a number of different bacterial strains, whereas in contrast complex **2.8** was selectively active against one strain. This implies that specific substitutions onto the aryl ring may lead to selective activity, while substitutions into the aryl ring (to create a pyridyl ring) lead to broad spectrum activity.

Table 5.11: Growth inhibition of cobalt(III) *tris*-picolinamide complexes against bacterial strains

Complex	Inhibition / %				
	E. coli	*K. pneumoniae*	*A. baumannii*	*P. aeruginosa*	*S. aureus*
2.1a	0.0	8.3	1.6	0.0	0.0
2.2	2.4	13.4	7.6	0.0	2.2
2.3	9.8	17.6	13.4	0.0	18.7
2.4	7.2	18.8	11.2	0.0	14.9
2.5	3.5	19.2	14.8	0.0	23.8
2.6	6.0	19.4	12.1	0.0	16.7
2.7	0.0	17.2	13.6	0.0	6.5
2.8	0.0	17.7	9.6	0.0	65.9
2.9	5.4	17.5	11.3	0.0	13.0
2.10	0.9	17.9	8.1	0.0	16.0
2.11	0.0	17.6	6.9	0.0	15.1
2.12	5.2	15.2	7.6	1.5	12.0
2.13	4.4	12.4	7.2	3.1	8.6
2.14a	8.7	18.2	10.3	0.0	18.6
2.15a	6.8	17.3	10.2	0.0	15.0
2.16a	0.4	18.1	13.1	0.0	20.9
2.17a	1.3	16.7	9.3	0.0	13.9
2.18a	1.3	19.6	8.5	0.0	14.8
2.1b	11.4	18.0	13.7	0.0	16.8
2.14b	4.3	19.2	13.1	0.0	14.2
2.15b	5.4	17.5	12.9	0.0	18.5
2.16b	6.3	17.7	10.7	0.0	15.4
2.17b	6.3	14.8	12.0	0.0	5.3
2.18b	4.3	15.8	9.5	0.0	6.9

Table 5.12: Growth inhibition of cobalt(II) *bis*-picolinamide complexes against bacterial strains

Complex	Inhibition / %				
	E. coli	*K. pneumoniae*	*A. baumannii*	*P. aeruginosa*	*S. aureus*
3.1	4.4	19.1	16.7	0.0	9.1
3.2	0.0	19.2	12.5	0.0	13.3
3.3	0.0	14.7	8.6	0.0	10.0
3.4	5.2	15.6	11.7	0.0	6.6
3.5	3.5	14.2	12.8	0.0	8.4
3.6	0.0	16.2	12.8	0.0	6.7
3.7	2.1	17.3	11.7	0.0	2.3
3.8	0.0	17.5	7.8	0.0	5.8
3.9	0.0	16.1	11.8	0.0	0.5
3.10	3.1	11.2	6.9	0.0	0.0

Table 5.13: Growth inhibition of cobalt(III) mixed ligand complexes against bacterial strains

Complex	Inhibition / %				
	E. coli	*K. pneumoniae*	*A. baumannii*	*P. aeruginosa*	*S. aureus*
4.2	0.0	13.1	4.7	0.0	0.0
4.3	0.0	8.5	0.0	0.0	0.0
4.4	0.0	10.0	1.6	0.0	0.0
4.5	0.0	11.7	0.0	0.0	3.9
4.6	0.0	12.1	7.1	0.0	0.5
4.7	0.0	2.2	0.0	0.0	0.9
4.8	0.0	4.2	0.0	0.0	0.0
4.9	0.0	2.0	0.0	0.0	1.1
4.10	0.0	0.0	0.0	0.0	0.0
4.11	0.0	0.0	0.0	0.0	4.6
4.12	0.0	0.0	0.0	0.0	2.7
4.13	0.0	5.0	0.0	0.0	0.0
4.14	0.0	8.1	0.0	0.0	0.0
4.15	0.0	5.4	0.0	0.0	0.0
4.16	0.0	3.5	0.0	0.0	4.2
4.17	0.0	3.1	0.0	6.0	0.0

5.6 Anti-fungal activity

The incidence of fungal infections has increased in recent years and these are often associated with significant mortality rates.[84] This is partly due to the increase in immunocompromised patients, for example patients with HIV or patients undergoing organ transplantation or aggressive chemotherapy, for which opportunistic fungal or bacterial infections are particularly worrisome.[85] Clinical outcomes are often poor and drug resistant strains of common organism such as *Candida albicans* are emerging.[86] In the USA, *C. albicans* is now the fourth most common iatrogenic bloodstream infection[87] and in developing healthcare systems

infection with fungi of the *Cryptococcus* genus causes more deaths in HIV patients than tuberculosis.[88] There are currently only four main classes of anti-fungal drugs: polyenes, azoles, allylamines and echinocandins; and thus there is a crucial requirement for the development of new anti-fungal drugs.[85]

Despite the need for new drugs, there has been relatively little investigation into the anti-fungal potential of organometallic or coordination complexes. There are only a few publications which examine the anti-fungal activity of cobalt complexes and these exclusively examine cobalt(II) complexes. Schiff base complexes, some based on amino acids, have shown promising activity against *C. albicans* or *Aspergillus* genus fungi or general broad spectrum activity.[89-92] Other ligands investigated include azoles,[93] cephalexins,[94] sulphonamides,[95] dithiones,[96] thiolenes,[97] diols[98] and 8-hydroxyquinoline[99] which tend to show broad spectrum activity against a variety of fungi. In cases where analogous copper(II) complexes are investigated, these show improved activity compared to the cobalt(II) complexes.[92, 98]

Complexes were screened for their anti-fungal activity against *Candida albicans* (*C. albicans*) and *Cryptococcus neoformans* (*C. neoformans*) by CO-ADD at The University of Queensland. The complexes, at a single concentration of 32 µg mL^{-1}, were incubated with the fungal strains at 35 °C for 24 hours without shaking. All growth inhibition assays were performed in duplicate. Growth inhibition of *C. albicans* was determined by measuring absorbance at 530 nm. Growth inhibition of *C. neoformans* was determined by measuring the difference in absorbance between 580 and 600 nm following the addition of resazurin and incubation at 35 °C for a further two hours. Complexes with growth inhibition values greater than 80 % are classed as active and complexes with growth inhibition values in the range 50 - 80 % are classed as partially active.

Anti-fungal activity screening results for cobalt(III) *tris*-picolinamide complexes, cobalt(II) *bis*-picolinamide complexes and cobalt(III) mixed ligand complexes are summarised in **Table 5.14**, **Table 5.15** and **Table 5.16** respectively. Active complexes are highlighted in green and partially active complexes are highlighted in orange. Results for active and partially active complexes are also displayed in **Figure 5.15**. Complexes **4.1**, **4.18** and **4.19** were insufficiently soluble in DMSO and cell culture medium used for the growth inhibition assays and therefore no results could be obtained.

Table 5.14: Growth inhibition of cobalt(III) *tris*-picolinamide complexes against fungal strains

Complex	Inhibition / %	
	C. albicans	*C. neoformans*
2.1a	0.8	0.0
2.2	1.0	0.0
2.3	3.3	0.0
2.4	1.7	0.0
2.5	2.1	0.0
2.6	4.2	0.0
2.7	1.9	0.0
2.8	2.2	0.0
2.9	0.8	0.0
2.10	3.6	0.0
2.11	2.6	0.0
2.12	0.8	0.0
2.13	9.7	0.0
2.14a	3.2	0.0
2.15a	1.4	0.0
2.16a	3.9	0.0
2.17a	0.8	0.0
2.18a	5.3	0.0
2.1b	5.3	0.0
2.14b	7.7	0.0
2.15b	6.6	0.0
2.16b	4.3	0.0
2.17b	6.7	7.7
2.18b	2.5	0.0

Table 5.15: Growth inhibition of cobalt(II) *bis*-picolinamide complexes against fungal strains

Complex	Inhibition / %	
	C. albicans	*C. neoformans*
3.1	86.8	0.0
3.2	81.0	0.0
3.3	85.9	0.0
3.4	78.6	0.0
3.5	72.8	0.0
3.6	73.5	0.0
3.7	63.5	0.0
3.8	72.0	0.0
3.9	44.0	0.0
3.10	26.0	0.0
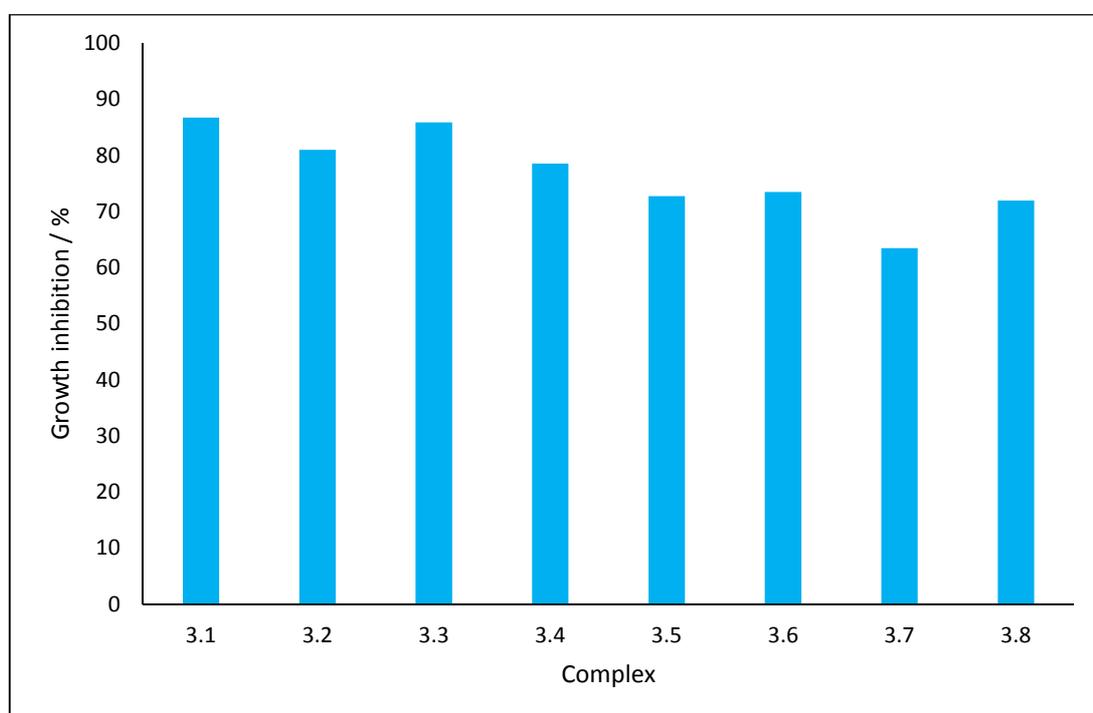
Figure 5.15: Growth inhibition of *C. albicans* by cobalt(II) *bis*-picolinamide thiocyanate complexes

Table 5.16: Growth inhibition of cobalt(III) mixed ligand complexes against fungal strains

Complex	Inhibition / %	
	C. albicans	*C. neoformans*
4.2	5.2	0.0
4.3	9.9	0.0
4.4	20.6	0.0
4.5	7.4	0.0
4.6	3.7	0.0
4.7	9.9	0.0
4.8	6.0	0.0
4.9	14.6	0.0
4.10	30.3	0.0
4.11	1.2	0.0
4.12	6.4	0.0
4.13	9.2	16.8
4.14	9.1	0.0
4.15	6.3	0.0
4.16	5.9	7.2
4.17	6.2	7.0

Only eight complexes displayed any notable activity against one of the fungal strains screened, all other complexes were inactive. All of the cobalt(II) *bis*-picolinamide thiocyanate complexes **3.1** - **3.8** were selectively active or partially active against *C. albicans*, with growth inhibition in the range 63.5 - 86.8 %. The analogous cobalt(II) complex with chlorides as the axial ligands (**3.9**) and the analogous dicationic cobalt(II) complex with aquas as the axial ligands (**3.10**) are inactive. The analogous cobalt(III) *tris*-picolinamide complexes are also inactive. Therefore, the axial thiocyanate ligands are essential for the anti-fungal activity of the complexes. Potentially, within the cell, the thiocyanate ligands are either released or exchanged, resulting in free thiocyanate ions which are known to be toxic.[100] In the saliva, salivary peroxidase catalyses the oxidation of thiocyanate (SCN⁻) by hydrogen peroxide to hypothiocyanite (NCSO⁻) and hypothiocyanous acid (NCSOH).[101, 102]

These compounds have been shown to inhibit the viability of *C. albicans*.[103] As toxicity to *C. albicans* is seen in these experiments, similar peroxidase enzymes within *C. albicans* may allow the formation of these toxic compounds leading to inhibition of growth. If these peroxidase enzymes are present at a higher concentration, or are more active or specific to *C. albicans* compared to *C. neoformans*, this may explain why these complexes are selectively toxic.

While all of the cobalt(II) *bis*-picolinamide thiocyanate complexes are at least partially active, the three active complexes (**3.1** - **3.3**) all contain the axial thiocyanates in the *trans* configuration; although the differences in percentage growth inhibition are fairly small. It is unknown if the positioning of the thiocyanate ligands has a causative effect on the anti-fungal activity. However, thiocyanate is a known *trans*-directing ligand[104] and different kinetics have been noted previously for *trans* and *cis* complexes with cobalt[105, 106] and a number of other metals.[107-109] Therefore, in the *trans* configuration the thiocyanates are released more readily, leading to greater concentrations of intracellular thiocyanate and its metabolites, hence a greater percentage of growth inhibition. Alternatively, the *cis* geometry may be more thermodynamically stable compared to the *trans* geometry, again favouring ligand substitution in the *trans* positions.[110]

Direct comparison of these results for cobalt picolinamide complexes with similar cobalt(II) Schiff base complexes in the literature is not possible due to use of different methods to quantify anti-fungal activity (percentage growth inhibition versus zone of inhibition). However, these Schiff base complexes display broad spectrum activity against a number of other fungal strains, whereas the cobalt picolinamide complexes show selectivity for *C. albicans*.[89, 90]

Complexes with sufficient activity (**3.1** - **3.7**) underwent hit confirmation to determine their minimum inhibitory concentration (MIC). MIC is the lowest drug concentration which prevents visible growth of the pathogen, in this case a fungus. The MIC values for complexes **3.1** - **3.7** were determined against *C. albicans* and *C. neoformans* by CO-ADD at The University of Queensland. The complexes, at eight concentrations, were incubated with the cell suspensions of the fungal strains at 35 °C for 36 hours without shaking. All growth inhibition assays were performed in duplicate. Growth inhibition of *C. albicans* was determined by measuring absorbance at 630 nm. Growth inhibition of *C. neoformans* was determined by measuring the difference in absorbance between 570 and 600 nm following the addition of

resazurin and incubation at 35 °C for a further two hours. The MIC was defined as the lowest concentration at which the growth was fully inhibited, set at ≥ 80% for *C. albicans* and inhibition ≥ 70% for *C. neoformans* (due to higher variance in growth and inhibition of *C. neoformans* compared to *C. albicans*). Complexes with MIC less than 16 µg ml^{-1} are classed as confirmed active hits. The cytotoxicity of these complexes was also determined against the HEK293 (human embryonic kidney) cell line. The complexes, at eight concentrations, were incubated with the cells at 37 °C for three hours in 5 % CO_2. Cytotoxicity was measured by fluorescence, with excitation at 560 nm and emission at 590 nm, following the addition of resazurin. Cytotoxicity is expressed in terms of CC_{50}, which is defined as the concentration of drug required to produce 50 % of cell death.

Anti-fungal MICs and cytotoxicity results for complexes **3.1** - **3.6** and **3.8** are summarised in **Table 5.17**. Despite displaying growth inhibition of *C. albicans* in the initial screen, none of the complexes displayed sufficient activity against *C. albicans* (MIC ≤ 16 µg ml^{-1}) to be classified as active hits. Perhaps the kinetics of ligand substitution resulting in free thiocyanate or the conversion of thiocyanate to hypothiocyanite is insufficiently rapid to allow sufficient anti-fungal activity. Complexes were also shown to be non-toxic to HEK293 cells, these results are in agreement with the previous cytotoxicity studies performed which also showed these complexes to be non-toxic to three different cell lines (**Chapter 5.1**).

Table 5.17: The anti-fungal toxicity and cytotoxicity of complexes **3.1** - **3.6** and **3.8**

Complex	MIC / µg ml^{-1}		CC_{50} / µg ml^{-1}
	C. albicans	*C. neoformans*	HEK293
3.1	> 32	> 32	> 32
3.2	> 32	> 32	> 32
3.3	> 32	> 32	> 32
3.4	> 32	> 32	> 32
3.5	> 32	> 32	> 32
3.6	> 32	> 32	> 32
3.8	> 32	> 32	> 32

5.7 Conclusions

Three series of cobalt picolinamide complexes were screened for their anti-cancer activity. Six complexes, mainly from the cobalt(III) *tris*-picolinamide series, were cytotoxic to pancreatic carcinoma, colon carcinoma and retinal epithelial cells. Within the cobalt(III) *tris*-picolinamide series, coordination isomers with *N,O* coordination were more active than the analogous *N,N* coordination complexes. This work represents the first time that coordination isomers of metal picolinamide complexes have been investigated and the first time that this trend in potency has been observed. Cobalt *tris*-picolinamide complexes were tested in a low oxygen (hypoxic) environment and found to be less active compared to in a normal oxygen (normoxic environment). Two complexes (**2.13** and **2.16b**) with high potency were designated as lead compounds and underwent mechanistic investigation. Both complexes inhibited cell proliferation rather than induced cell death at physiologically relevant concentrations and were found to act through interruption of M phase of the cell cycle. Both lead complexes were active against cancer stem cell enriched breast cancer cells and were able to inhibit mammosphere formation and viability.

The three series of cobalt picolinamide complexes were also screened for their anti-bacterial and anti-fungal activity. Complex **2.8** showed selective activity against the bacterium *S. aureus*, but all other complexes were inactive. All of the cobalt(II) *bis*-picolinamide thiocyanate complexes were selectively active against the fungus *C. albicans*. The thiocyanate axial ligands are essential for toxicity, possibly due to their ability to undergo ligand substitution to release free thiocyanate intracellularly. However, despite activity against *C. albicans* in the primary anti-fungal screen, the MIC values for these complexes were deemed insufficiently active to define the cobalt(II) *bis*-picolinamide thiocyanate complexes as active hits or to continue further study.

5.8 References

1.   E. L. Andrade, A. F. Bento, J. Cavalli, S. K. Oliveira, C. S. Freitas, R. Marcon, R. C. Schwanke, J. M. Siqueira and J. B. Calixto, *Brazilian Journal of Medical and Biological Research*, 2016, **49**.

2.   M. M. Hann, A. R. Leach and G. Harper, *Journal of Chemical Information and Computer Sciences*, 2001, **41**, 856-864.

3.   J. P. Hughes, S. Rees, S. B. Kalindjian and K. L. Philpott, *British Journal of Pharmacology*, 2011, **162**, 1239-1249.

4.   F. E. Koehn and G. T. Carter, *Nature Reviews Drug Discovery*, 2005, **4**, 206-220.

5.   T. L. Riss, R. A. Moravec, A. L. Niles, S. Duellman, H. A. Benink, T. J. Worzella and L. Minor, in *Assay guidance manual*, Editon edn., 2013.

6.   T. Mosmann, *Journal of Immunological Methods*, 1983, **65**, 55-63.

7.   J. C. Stockert, A. Blazquez-Castro, M. Canete, R. W. Horobin and A. Villanueva, *Acta Histochemica*, 2012, **114**, 785-796.

8.   C. Bresson, C. Darolles, A. Carmona, C. Gautier, N. Sage, S. Roudeau, R. Ortega, E. Ansoborlo and V. Malard, *Metallomics*, 2013, **5**, 133-143.

9.   Y. M. Kwon, Z. Xia, S. Glyn-Jones, D. Beard, H. S. Gill and D. W. Murray, *Biomedical Materials*, 2009, **4**.

10.   T. Sakagami, K. Satoh, M. Ishihara, H. Sakagami, F. Takeda, M. Kochi and M. Takeda, *Anticancer Research*, 2000, **20**, 3143-3150.

11.   B. Scharf, C. C. Clement, V. Zolla, G. Perino, B. Yan, S. G. Elci, E. Purdue, S. Goldring, F. Macaluso, N. Cobelli, R. W. Vachet and L. Santambrogio, *Scientific Reports*, 2014, **4**.

12.   Z. Almodares, University of Leeds, 2010.

13.   S. H. van Rijt, A. J. Hebden, T. Amaresekera, R. J. Deeth, G. J. Clarkson, S. Parsons, P. C. McGowan and P. J. Sadler, *Journal of Medicinal Chemistry*, 2009, **52**, 7753-7764.

14.   G. M. Cooper, *The cell: a molecular approach*, Sinauer Associates, 2000.

15.   H. Lodish, A. Berk, S. L. Zipursky, D. Baltimore and J. Darnell, *Molecular cell biology*, Fourth edn., W. H. Freeman, 2000.

16.   J. C. Swarts, T. G. Vosloo, S. J. Cronje, W. C. Du Plessis, C. E. J. Van Rensbur, E. Kreft and J. E. Van Lier, *Anticancer Research*, 2008, **28**, 2781-2784.

17.   G. Gasser, I. Ott and N. Metzler-Nolte, *Journal of Medicinal Chemistry*, 2011, **54**, 3-25.

18.   C. Ornelas, *New Journal of Chemistry*, 2011, **35**, 1973-1985.

19.   N. K. Kaushik, A. Mishra, A. Ali, J. S. Adhikari, A. K. Verma and R. Gupta, *Journal of Biological Inorganic Chemistry*, 2012, **17**, 1217-1230.

20.   J. A. Bertout, S. A. Patel and M. C. Simon, *Nature Reviews Cancer*, 2008, **8**, 967-975.

21.   P. H. Maxwell, C. W. Pugh and P. J. Ratcliffe, *Current Opinion in Genetics & Development*, 2001, **11**, 293-299.

22.   C. Michiels, *American Journal of Pathology*, 2004, **164**, 1875-1882.

23.   A. Weidemann and R. S. Johnson, *Cell Death and Differentiation*, 2008, **15**, 621-627.

24.   J. E. Ziello, I. S. Jovin and Y. Huang, *Yale Journal of Biology and Medicine*, 2007, **80**, 51-60.

25.   C. J. Hu, L. Y. Wang, L. A. Chodosh, B. Keith and M. C. Simon, *Molecular and Cellular Biology*, 2003, **23**, 9361-9374.

26.   G. L. Semenza, in *Annual Review of Pathology: Mechanisms of Disease, Vol 9*, eds. A. K. Abbas, S. J. Galli and P. M. Howley, Editon edn., 2014, vol. 9, pp. 47-71.

27.   H. Zhong, A. M. De Marzo, E. Laughner, M. Lim, D. A. Hilton, D. Zagzag, P. Buechler, W. B. Isaacs, G. L. Semenza and J. W. Simons, *Cancer Research*, 1999, **59**, 5830-5835.

28.   I. F. Tannock, *Lancet*, 1998, **351**, 9-16.

29.   J. M. Brown and W. R. William, *Nature Reviews Cancer*, 2004, **4**, 437-447.

30.   K. M. Comerford, T. J. Wallace, J. Karhausen, N. A. Louis, M. C. Montalto and S. P. Colgan, *Cancer Research*, 2002, **62**, 3387-3394.

31.   N. Graf and S. J. Lippard, *Advanced Drug Delivery Reviews*, 2012, **64**, 993-1004.

32.   M. D. Hall and T. W. Hambley, *Coordination Chemistry Reviews*, 2002, **232**, 49-67.

33.   C. G. Hartinger, M. A. Jakupec, S. Zorbas-Seifried, M. Groessl, A. Egger, W. Berger, H. Zorbas, P. J. Dyson and B. K. Keppler, *Chemistry & Biodiversity*, 2008, **5**, 2140-2155.

34.   A. Levina, A. Mitra and P. A. Lay, *Metallomics*, 2009, **1**, 458-470.

35.   L. L. Parker, S. M. Lacy, L. J. Farrugia, C. Evans, D. J. Robins, C. C. O'Hare, J. A. Hartley, M. Jaffar and I. J. Stratford, *Journal of Medicinal Chemistry*, 2004, **47**, 5683-5689.

36.   T. W. Failes and T. W. Hambley, *Journal of Inorganic Biochemistry*, 2007, **101**, 396-403.

37.   C. R. Munteanu and K. Suntharalingam, *Dalton Transactions*, 2015, **44**, 13796-13808.

38.   B. A. Teicher, M. J. Abrams, K. W. Rosbe and T. S. Herman, *Cancer Research*, 1990, **50**, 6971-6975.

39.   D. C. Ware, B. G. Siim, K. G. Robinson, W. A. Denny, P. J. Brothers and G. R. Clark, *Inorganic Chemistry*, 1991, **30**, 3750-3757.

40.   S. Kummar, M. Gutierrez, J. H. Doroshow and A. J. Murgo, *British Journal of Clinical Pharmacology*, 2006, **62**, 15-26.

41.   A. Sparreboom, M. J. A. de Jonge and J. Verweij, *European Journal of Cancer*, 2002, **38**, 18-22.

42.   M. V. Blagosklonny and A. B. Pardee, *Cell Cycle*, 2002, **1**, 103-110.

43.   S. Lapenna and A. Giordano, *Nature Reviews Drug Discovery*, 2009, **8**, 547-566.

44.   S. Payne and D. Miles, in *Scott-Brown's Otorhinolaryngology: Head and Neck Surgery*, CRC Press, Editon edn., 2008.

45.   M. A. Shah and G. K. Schwartz, *Clinical Cancer Research*, 2001, **7**, 2168-2181.

46.   G. I. Shapiro and J. W. Harper, *Journal of Clinical Investigation*, 1999, **104**, 1645-1653.

47.   Biology Notes Help, *Cell cycle regulation*, http://www.biologynoteshelp.com/mpfapccyclosome/, Accessed 08/01/2017.

48.   American Chemical Society, *Chemotherapy drugs: how they work*, http://www.cancer.org/acs/groups/cid/documents/webcontent/002995-pdf.pdf, Accessed 08/01/2017.

49.   M. C. Isoldi, M. A. Visconti and A. M. D. Castrucci, *Mini-Reviews in Medicinal Chemistry*, 2005, **5**, 685-695.

50.   ChemoMetec   A/S,   *Two-step   cell   cycle   analysis*,
        https://chemometec.com/wp-content/uploads/2015/03/994-3001-Two-
        Step-Cell-Cycle-Assay.pdf, Accessed 02/01/2017.

51.   S. Chatterjee, S. Kundu, A. Bhattacharyya, C. G. Hartinger and P. J. Dyson,
        Journal of Biological Inorganic Chemistry, 2008, **13**, 1149-1155.

52.   L. He, S. Y. Liao, C. P. Tan, R. R. Ye, Y. W. Xu, M. Zhao, L. N. Ji and Z. W. Mao,
        Chem.-Eur. J., 2013, **19**, 12152-12160.

53.   K. S. M. Smalley, R. Contractor, N. K. Haass, A. N. Kulp, G. E. Atilla-Gokcumen,
        D. S. Williams, H. Bregman, K. T. Flaherty, M. S. Soengas, E. Meggers and M.
        Herlyn, *Cancer Research*, 2007, **67**, 209-217.

54.   A. Wieczorek, A. Blauz, J. Zakrzewski, B. Rychlik and D. Plazuk, *ACS Medicinal
        Chemistry Letters*, 2016, **7**, 612-617.

55.   P. B. Gupta, C. L. Chaffer and R. A. Weinberg, *Nature Medicine*, 2009, **15**,
        1010-1012.

56.   D. Hanahan and R. A. Weinberg, *Cell*, 2011, **144**, 646-674.

57.   L. V. Nguyen, R. Vanner, P. Dirks and C. J. Eaves, *Nature Reviews Cancer*, 2012,
        12, 133-143.

58.   D. R. Pattabiraman and R. A. Weinberg, *Nature Reviews Drug Discovery*, 2014,
        13, 497-512.

59.   J. N. Rich, *Cancer Research*, 2007, **67**, 8980-8984.

60.   K. Chen, Y. H. Huang and J. L. Chen, *Acta Pharmacologica Sinica*, 2013, **34**,
        732-740.

61.   A. Eramo, F. Lotti, G. Sette, E. Pilozzi, M. Biffoni, A. Di Virgilio, C. Conticello, L.
        Ruco, C. Peschle and R. De Maria, *Cell Death and Differentiation*, 2008, **15**,
        504-514.

62.   R. Lamb, H. Harrison, J. Hulit, D. L. Smith, M. P. Lisanti and F. Sotgia,
        Oncotarget, 2014, **5**, 11029-11037.

63.   X. Y. Ning, J. C. Shu, Y. Q. Du, Q. W. Ben and Z. S. Li, *Cancer Biology & Therapy*,
        2013, **14**, 295-303.

64.   J. Kaiser, *Science*, 2015, **347**, 226-229.

65.   P. B. Cressey, A. Eskandari, P. M. Bruno, C. X. Lu, M. T. Hemann and K.
        Suntharalingam, *Chembiochem*, 2016, **17**, 1713-1718.

66.   J. N. Boodram, I. J. McGregor, P. M. Bruno, P. B. Cressey, M. T. Hemann and K. Suntharalingam, *Angewandte Chemie-International Edition*, 2016, **55**, 2845-2850.

67.   A. Eskandari, J. N. Boodram, P. B. Cressey, C. X. Lu, P. M. Bruno, M. T. Hemann and K. Suntharalingam, *Dalton Transactions*, 2016, **45**, 17867-17873.

68.   M. Gonzalez-Bartulos, C. Aceves-Luquero, J. Qualai, O. Cusso, M. A. Martinez, S. F. de Mattos, J. A. Menendez, P. Villalonga, M. Costas, X. Ribas and A. Massaguer, *Plos One*, 2015, **10**.

69.   E. Kioseoglou, S. Petanidis, C. Gabriel and A. Salifoglou, *Coordination Chemistry Reviews*, 2015, **301**, 87-105.

70.   T. T. Zou, C. T. Lum, C. N. Lok, J. J. Zhang and C. M. Che, *Chemical Society Reviews*, 2015, **44**, 8786-8801.

71.   K. Suntharalingam, W. Lin, T. C. Johnstone, P. M. Bruno, Y. R. Zheng, M. T. Hemann and S. J. Lippard, *Journal of the American Chemical Society*, 2014, **136**, 14413-14416.

72.   P. B. Gupta, T. T. Onder, G. Z. Jiang, K. Tao, C. Kuperwasser, R. A. Weinberg and E. S. Lander, *Cell*, 2009, **138**, 645-659.

73.   G. Dontu, W. M. Abdallah, J. M. Foley, K. W. Jackson, M. F. Clarke, M. J. Kawamura and M. S. Wicha, *Genes & Development*, 2003, **17**, 1253-1270.

74.   World Health Organisation, *Antimicrobial resistance*, http://www.who.int/mediacentre/factsheets/fs194/en/, Accessed 20/12/2016.

75.   H. W. Boucher, G. H. Talbot, J. S. Bradley, J. E. Edwards, D. Gilbert, L. B. Rice, M. Scheld, B. Spellberg and J. Bartlett, *Clinical Infectious Diseases*, 2009, **48**, 1-12.

76.   J. N. Pendleton, S. P. Gorman and B. F. Gilmore, *Expert Review of Anti-Infective Therapy*, 2013, **11**, 297-308.

77.   S. Santajit and N. Indrawattana, *Biomed Research International*, 2016.

78.   C. J. H. von Wintersdorff, J. Penders, J. M. van Niekerk, N. D. Mills, S. Majumder, L. B. van Alphen, P. H. M. Savelkoul and P. F. G. Wolffs, *Frontiers in Microbiology*, 2016, **7**.

79.   J. M. Conly and B. L. Johnston, *Canadian Journal of Infectious Diseases and Medical Microbiology*, 2005, **16**, 159-160.

80.   C. Lee Ventola, *Pharmacy and Therapeutics*, 2015, **40**, 277-283.

81.   F. P. Dwyer, E. C. Gyarfas, W. P. Rogers and J. H. Koch, *Nature*, 1952, **170**, 190-191.

82.   E. L. Chang, C. Simmers and D. A. Knight, *Pharmaceuticals*, 2010, **3**, 1711-1728.

83.   A. Mishra, N. K. Kaushik, A. K. Verma and R. Gupta, *European Journal of Medicinal Chemistry*, 2008, **43**, 2189-2196.

84.   J. R. Perfect, *Expert Opinion on Emerging Drugs*, 2016, **21**, 129-131.

85.   V. T. Andriole, *Journal of Antimicrobial Chemotherapy*, 1999, **44**, 151-162.

86.   G. D. Brown, D. W. Denning, N. A. R. Gow, S. M. Levitz, M. G. Netea and T. C. White, *Science Translational Medicine*, 2012, **4**.

87.   R. E. Lewis, *Current Medical Research and Opinion*, 2009, **25**, 1732-1740.

88.   B. J. Park, K. A. Wannemuehler, B. J. Marston, N. Govender, P. G. Pappas and T. A. Chiller, *Aids*, 2009, **23**, 525-530.

89.   S. Belaid, A. Landreau, S. Djebbar, O. Benali-Baitich, M. A. Khan and G. Bouet, *Transition Metal Chemistry*, 2008, **33**, 511-516.

90.   Z. H. Chohan, M. Arif, M. A. Akhtar and C. T. Supuran, *Bioinorganic Chemistry and Applications*, 2006.

91.   M. I. Khan, A. Khan, I. Hussain, M. A. Khan, S. Gul, M. Iqbal, R. Inayat Ur and F. Khuda, *Inorganic Chemistry Communications*, 2013, **35**, 104-109.

92.   P. V. Rao, S. Ammanni and S. Kalidasu, *E-Journal of Chemistry*, 2011, **8**, 470-478.

93.   M. Hanif and Z. H. Chohan, *Applied Organometallic Chemistry*, 2013, **27**, 36-44.

94.   Z. H. Chohan, H. Pervez, K. M. Khan, A. Rauf, G. M. Maharvi and C. T. Supuran, *Journal of Enzyme Inhibition and Medicinal Chemistry*, 2004, **19**, 85-90.

95.   Z. H. Chohan, H. A. Shad, M. H. Youssoufi and T. Ben Hadda, *European Journal of Medicinal Chemistry*, 2010, **45**, 2893-2901.

96.   Z. H. Chohan, *Applied Organometallic Chemistry*, 2006, **20**, 112-116.

97.   Z. H. Chohan, A. U. Shaikh and C. T. Supuran, *Journal of Enzyme Inhibition and Medicinal Chemistry*, 2006, **21**, 733-740.

98.   N. V. Loginova, T. V. Koval'chuk, G. I. Polozov, N. P. Osipovich, P. G. Rytik, Kucherov, II, A. A. Chernyavskaya, V. L. Sorokin and O. I. Shadyro, *European Journal of Medicinal Chemistry*, 2008, **43**, 1536-1542.

99.   M. A. El-Gahami, Z. A. Khafagy, A. M. M. Ali and N. M. Ismail, *Journal of Inorganic and Organometallic Polymers*, 2004, **14**, 117-129.

100.   S. J. Nessim and R. M. A. Richardson, *Asaio Journal*, 2006, **52**, 479-481.

101.   M. T. Ashby, *Journal of Dental Research*, 2008, **87**, 900-914.

102.   A. Slungaard and J. R. Mahoney, *Journal of Biological Chemistry*, 1991, **266**, 4903-4910.

103.   M. Lenander-Lumikari, *Oral Microbiology and Immunology*, 1992, **7**, 315-320.

104.   C. E. Housecroft and A. G. Sharpe, *Inorganic Chemistry*, Fourth edn., Pearson Ltd., 2012.

105.   J. V. Quagliano and L. Schubert, *Chemical Reviews*, 1952, **50**, 201-260.

106.   J. K. Yandell and L. A. Tomlins, *Australian Journal of Chemistry*, 1978, **31**, 561-571.

107.   J. L. Burmeister and J. B. Melpolder, *Journal of the Chemical Society-Chemical Communications*, 1973, 613-614.

108.   W. R. Bushey and J. H. Espenson, *Inorganic Chemistry*, 1977, **16**, 2772-2776.

109.   K. Kondo, H. Nishio and F. Nakashio, *Journal of Chemical Engineering of Japan*, 1990, **23**, 236-238.

110.   M. Dakovic, D. Vila-Vicosa, M. J. Calhorda and Z. Popovic, *CrystEngComm*, 2011, **13**, 5863-5871.

Chapter 6

Chemical investigation of cobalt picolinamide complexes

6. Chemical investigation

After the identification of promising (lead) drugs during initial biological screening it is necessary to investigate aspects such as the distribution of the drug within the body, the drug metabolism and any drug toxicity to non-target cells.[1, 2] These in depth investigations are essential to identify the potential pitfalls or adverse effects of new drugs. Several promising leads fail at this stage of development e.g. the titanium containing anti-cancer drug budotitane which was removed from clinical trials due to problems with solubility, stability and non-target toxicity.[3-6] If such problems can be identified in the preclinical phase, they can inform further rounds of lead development and optimisation. The understanding of the relationship between the molecular structure of a drug and activity through structure-activity relationships (SAR) allows the modification of drugs to improve various biologically relevant characteristics.[7-9]

6.1 Hydrolysis studies

The proposed mechanism of action for several metal based anti-cancer drugs, including cisplatin, involves activation by hydrolysis.[10, 11] Initially such drugs are inert when administered and do not undergo ligand substitution, but upon transfer across the cell membrane, the change in chloride concentration permits ligand substitution reactions to occur leading to activation of the drug.[12-14] In other metal picolinamide complexes, the rate of hydrolysis was related to the cytotoxicity.[15] For example, within the McGowan group a greater degree of hydrolysis of ruthenium(III) *bis*-picolinamide dichloride complexes was associated with improved cytotoxicity whereas a lesser degree of hydrolysis of ruthenium(III) *bis*-picolinamide diiode complexes was associated with improved cytotoxicity.[16] Possible hydrolysis pathways for the cobalt picolinamide complexes involve the formation of cationic monoaqua or diaqua intermediates under physiological conditions (**Figure 6.1**).[17-20]
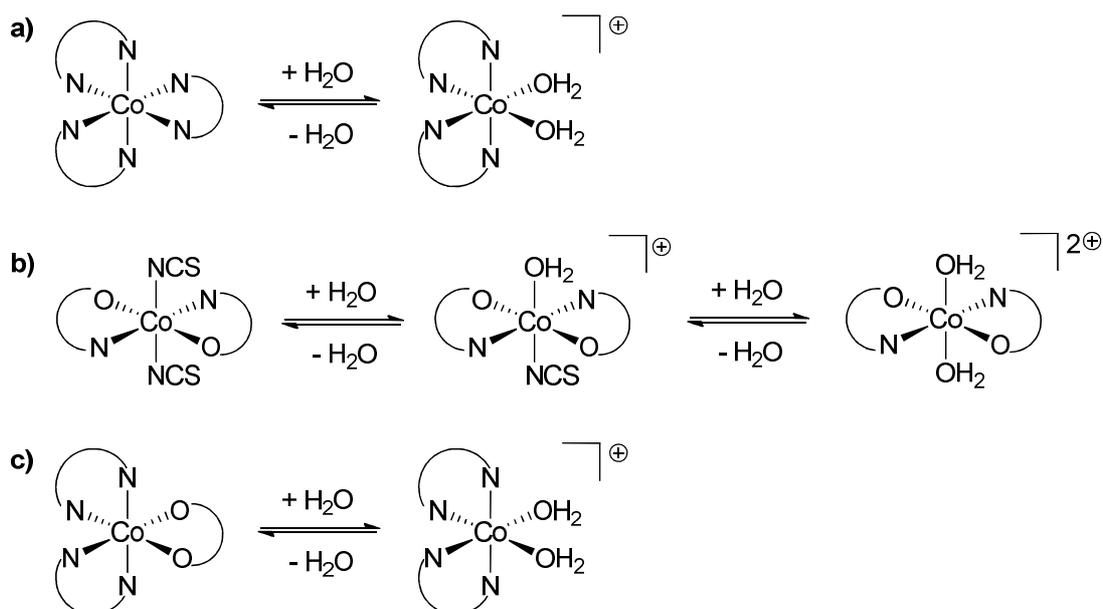
Figure 6.1: Potential hydrolysis pathways for cobalt picolinamide complexes

Hydrolysis studies were performed using both ^{1}H NMR and UV/vis spectroscopy. NMR samples were prepared in 4:1 d_3-acetonitrile/D_2O to give a final concentration of 8 mg ml^{-1}. UV/vis samples were prepared in 4:1 acetonitrile/water to give a final concentration of 50 μM. Attempts to increase the concentration of water in these samples resulted in poor solubility. Samples were analysed using NMR and UV/vis spectroscopy every 24 hours over a period of five days, to correlate with the MTT assay. Following the final analyses, samples were analysed by electrospray mass spectrometry.

Hydrolysis was not investigated for all complexes; only for complexes with activity or partial activity against cancer cell lines (**2.13**, **2.16b** and **4.10**), their closest analogous inactive complex (**2.16a** and **4.11**) and an example of an unsubstituted complex from the series of cobalt picolinamide complexes synthesised (**2.1a**, **2.1b**, **4.3** and **4.13**). Cobalt(II) *bis*-picolinamide complexes (**3.1** - **3.10**) were insufficiently soluble in the acetonitrile/water mixture used in these investigations and therefore no results could be obtained. The changes in wavelengths of absorption bands from the UV/vis spectra are summarised in **Table 6.1** and UV/vis spectra of complexes which undergo changes over time are displayed in **Figure 6.2**. All complexes are stable in acetonitrile over the same time period with no concurrent changes to the NMR, UV/vis or mass spectra over time.

Table 6.1: Wavelengths of absorption bands of cobalt picolinamide complexes

Complex	Wavelength / nm	
	Day 0	Day 5
2.1a	204, 263, 298 (sh)	204, 263, 298 (sh)
2.13	207, 256, 299 (sh)	207, 256, 299 (sh)
2.16a	205, 262, 303 (sh)	205, 262, 303 (sh)
2.1b	202, 266, 300 (sh), 355 (sh)	202, 266, 300 (sh), 355 (sh)
2.16b	206, 256 (sh), 340 (sh)	206, 256 (sh), 340 (sh)
4.3	202, 266, 287 (sh), 354	202, 266, 287 (sh), 354
4.10	204, 258, 293, 369 (sh), 522	200, 231 (sh), 272
4.11	201, 259, 302, 384 (sh), 527	200, 229 (sh), 273
4.13	203, 267, 299, 373, 511	203, 270, 388 (sh), 419 (sh)

There is no change in the UV/vis, NMR or mass spectra for complexes **2.1a**, **2.13**, **2.16a**, **2.1b**, **2.16b** or **4.3** over the five day time period. This shows that the cobalt(III) *tris*-picolinamide complexes and cobalt(III) mixed ligand complexes with β-diketonate ligands are stable in solution and do not undergo hydrolysis. There is no difference in the degree of hydrolysis between the active and inactive complexes (from the same and different cobalt picolinamide series) and therefore the differences in anti-cancer activity are not due to differences in either the degree or rate of hydrolysis. This is in contrast to previously studied ruthenium(III) picolinamide complexes where hydrolysis was correlated with activity,[15, 16] the implication being that the mechanism of action of these complexes is different to those previously studied. This lack of a requirement for hydrolysis for activation of the drug is also different to a number of well-established and promising metal anti-cancer drugs such as cisplatin, KP1019 and NAMI-A.[10, 11, 21] However, the hydrolytic activation of these drugs has disadvantages; for example the rapid hydrolysis of cisplatin leads to reduced bioavailability due to binding to thiol containing proteins which is thought to be a key mechanism in cisplatin toxicity to non-target cells.[22-24] It is possible that the difference in mechanism for these cobalt picolinamide complexes, which do not seem to require hydrolytic activation, will limit some of these potential drug pitfalls.

Complex **4.10**   Complex **4.11**   Complex **4.13**
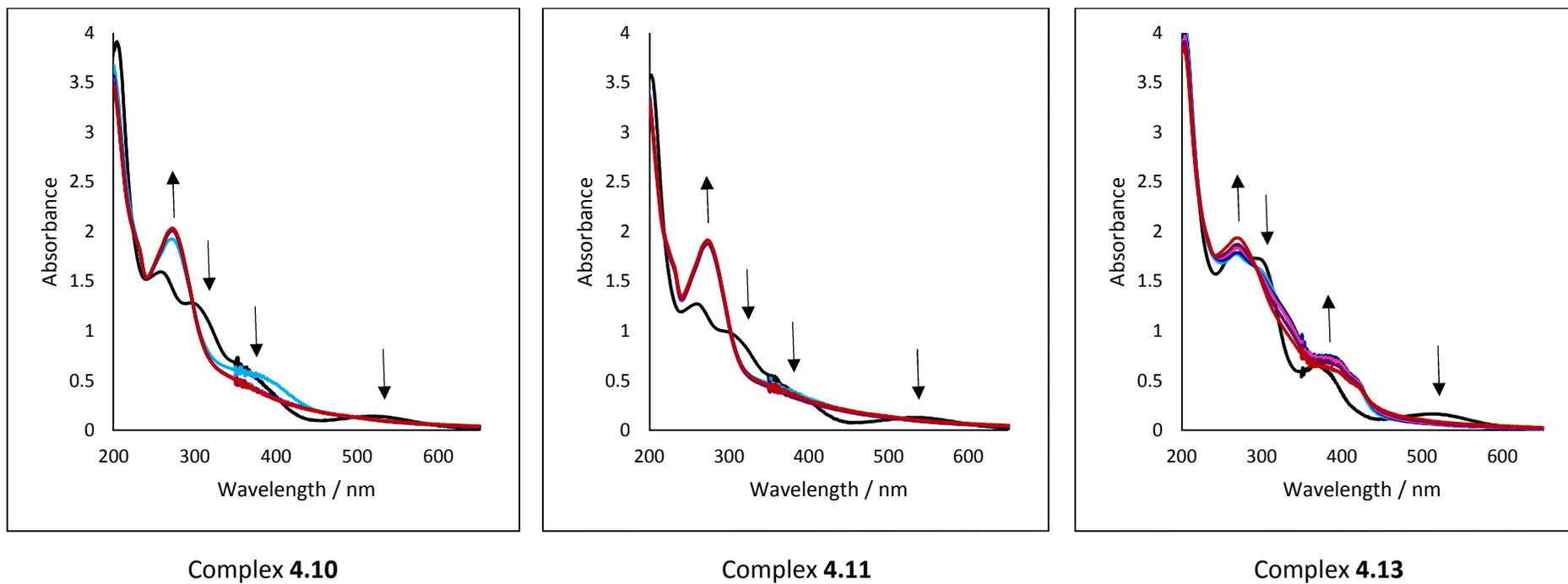
Figure 6.2: UV/vis spectra for the hydrolysis of complexes **4.10**, **4.11** and **4.13**

Key: Black = day 0, pale blue = day 1, dark blue = day 2, pink = day 3, purple = day 4, dark red = day 5

Distinct changes in the UV/vis spectra for complexes **4.10**, **4.11** and **4.13** are observed over the five day time period. These spectral changes are associated with changes in the colour of the samples themselves; the UV/vis solutions changing from red/purple on day 0 to brown by day 5. This shows that the cobalt(III) mixed ligand complexes with ferrocenyl β-diketonate ligands undergo hydrolysis over time. This susceptibility to hydrolysis is due to the presence of the ferrocenyl group as in the absence of ferrocene no hydrolysis is observed. On day 0, these complexes show an intense ligand based absorbance (π-π*) at approximately 200 nm with three less intense ligand based charge-transfer bands at 255 - 270 nm, 290 - 305 nm and 370 - 385 nm and finally a weak d-d transition at 510 - 530 nm. By day 5, the intense absorbance at around 200 nm remains but all of the weaker charge-transfer bands and d-d transition have disappeared and instead a distinct charge-transfer band at 270 - 275 nm has appeared. The spectra seen at day 5 are remarkably similar to the UV/vis spectra of the cobalt *bis*-picolinamide complexes, which also display an intense ligand based absorbance at around 200 nm and a distinct charge-transfer band at 275 - 285 nm. This suggests that during the hydrolysis experiment, the ferrocenyl β-diketonate ligand dissociates from the complex resulting in a cobalt *bis*-picolinamide complex with the axial ligands either aqua or acetonitrile molecules (**Figure 6.1c**).

Relative hydrolysis rates (**4.11** > **4.10** > **4.13**) can be inferred from the UV/vis spectra as by day 1 complex **4.11** is completely converted, by day 2 complex **4.10** is completely converted and by day 5 complex **4.13** is still undergoing conversion. Complex **4.11** in particular undergoes rapid hydrolysis as the colour change of the UV/vis solution is observed within 1 - 2 hours, and with only 20 % water component in the solution. The relative hydrolysis rates correlate with the ferrocenyl β-diketonate substituents present in the complexes. Complex **4.11** contains the most electron withdrawing substituent (CF_3) and has the most rapid hydrolysis rate, complex **4.10** contains a slightly less electron withdrawing substituent (CHF_2) and has a rapid hydrolysis rate and complex **4.13** contains an electron donating substituent (Ph) and has the slowest rate of hydrolysis.

The changes to the NMR and mass spectra during hydrolysis can be used to uncover the fate of complexes **4.10**, **4.11** and **4.13**. Example NMR spectra over time are shown for complex **4.11** (**Figure 6.3**) and the changes in the mass spectra for complexes **4.10**, **4.11** and **4.13** over time are summarised in **Table 6.2**.
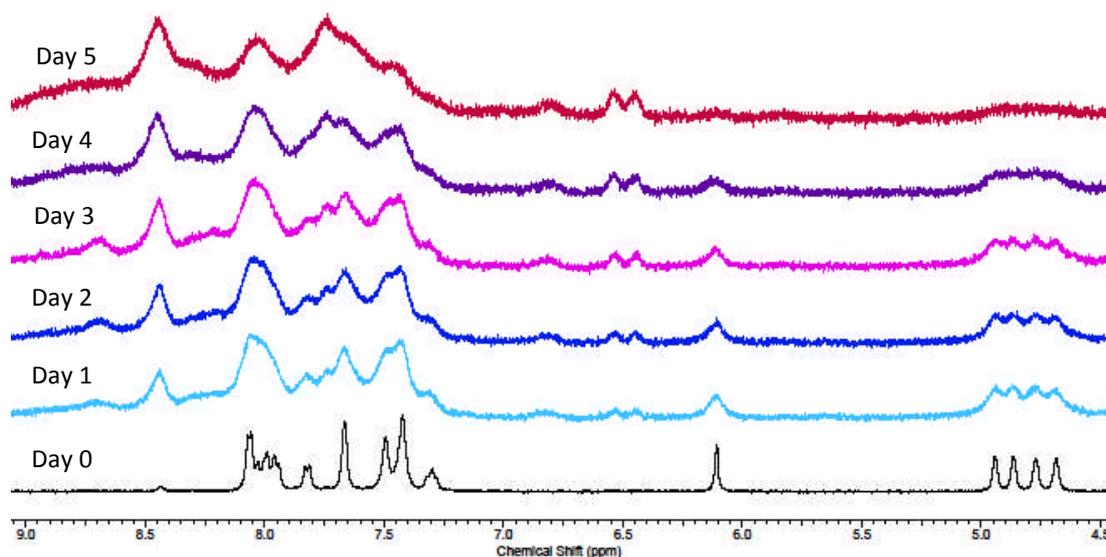
Figure 6.3: ^{1}H NMR spectra of complex **4.11** over time (4:1 CD$_3$CN/D$_2$O)

Table 6.2: Mass spectrometry peaks and assignments for complexes **4.10**, **4.11** and **4.13.** pica = picolinamide, keto = β-diketonate

Complex	Day 0		Day 5	
	Peak	**Assignment**	**Peak**	**Assignment**
4.10	1031.04 (100 %)	[Co(pica)$_2$(keto)]$^+$	725.02 (5 %)	[Co(pica)$_2$]$^+$
			335.06 (95 %)	pica$^+$
4.11	1049.04 (100 %)	[Co(pica)$_2$(keto)]$^+$	725.02 (5 %)	[Co(pica)$_2$]$^+$
			335.06 (95 %)	pica$^+$
4.13	1057.08 (100 %)	[Co(pica)$_2$(keto)]$^+$	725.02 (5 %)	[Co(pica)$_2$]$^+$
			335.06 (95 %)	pica$^+$

In the NMR spectra, complexes **4.10**, **4.11** and **4.13** all show broadening of peaks over time, revealing that either a charged or a paramagnetic species is being produced in solution. This could be consistent with the substitution of one of the bidentate ligands for two water molecules producing a cationic species (**Figure 6.1c**). In addition, the methine β-diketonate peak at 6.11 ppm disappears over time and new peaks at 6.45, 6.54, 6.80 and 8.45 ppm appear (**Figure 6.3**). The loss of the methine peak may indicate the dissociation of the ferrocenyl β-diketonate ligand from the complex with the new peaks either the result of the formation of a cobalt *bis*-picolinamide complex or degradation of the cobalt picolinamide complex.[18] This

supports the evidence from the UV/vis spectra, which suggests the formation of a cobalt *bis*-picolinamide complex. The new peaks at 6.45 - 6.80 may be explained by the shifting and splitting of the methine peak if new species are formed in solution from the free ferrocenyl β-diketonate ligands. The formation of new species in solution is supported by the fact that the ferrocenyl β-diketonate ligand with a trifluoromethyl group was cytotoxic as the free ligand.[25] But as the complex containing this ligand is non-cytotoxic the implication is that any ferrocenyl β-diketonate ligand released is either released in a modified form or can rapidly form new species in solution.

The mass spectra of complexes **4.10**, **4.11** and **4.13** also show changes over time. Initially the major peak is for the intact cobalt mixed ligand complex, but by day 5 the major peaks can be assigned to free picolinamide ligand and a cobalt *bis*-picolinamide complex. This supports evidence from NMR spectroscopy which suggests that the ferrocenyl β-diketonate ligand is substituted during hydrolysis. Unfortunately, no cobalt aqua species could be identified from the mass spectra which may imply that the complexes may simply undergo degradation rather than aquation over time.

In order to probe the behaviour of the complexes in solution, the ferrocenyl β-diketonate ligand component of complex **4.11** was studied in isolation under the same conditions as previously described (**Figure 6.4**). The UV/vis spectra showed distinct changes over time but the absorption bands observed did not correlate with complex **4.11**. The NMR spectra also showed peak broadening over time with the disappearance of the methine peak and appearance of two new peaks at 6.45 and 6.53 ppm, which may be due to shifting and splitting of the methine peak. The mass spectra also showed changes over time but none of the peaks after day 0 could be assigned due to the complex nature of the spectra. The ligand is stable in acetonitrile over the same time period with no concurrent changes to the NMR, UV/vis or mass spectra over time.[26]
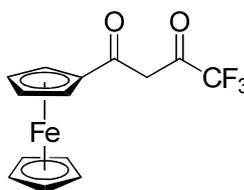
Figure 6.4: Ferrocenyl β-diketonate ligand component of complex **4.11**

During these hydrolysis experiments, dark brown crystals formed in the NMR tube containing the ferrocenyl β-diketonate ligand in 4:1 d_3-acetonitrile/D_2O. This hydrolysis product crystallised in a monoclinic cell and structural solutions were performed in the P 2_1/c space group with a single molecule in the asymmetric unit (**Figure 6.5**).
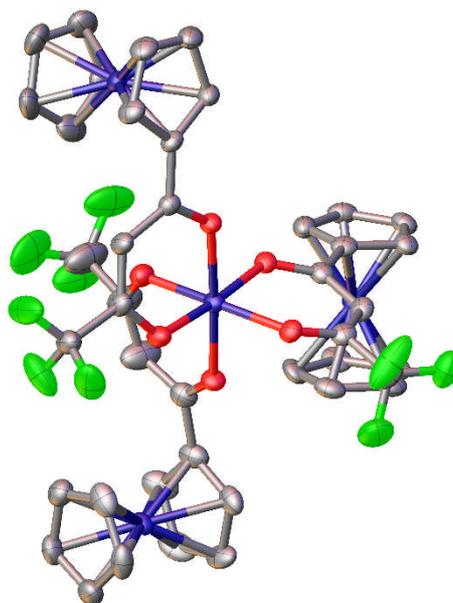
Figure 6.5: Molecular structure of the ferrocenyl β-diketonate ligand hydrolysis product. Displacement ellipsoids are at the 50 % probability level. Hydrogen atoms are omitted for clarity

This hydrolysis product is a metal complex with a central iron(III) surrounded by three ferrocenyl β-diketonate ligands (**Figure 6.6**). This is a surprising result as ferrocene has high stability and previous ferrocenyl β-diketonate ligands have proven stable in solution, however these were not investigated in water or for extended time periods.[27-30] The iron at the centre of this complex can only originate from the ferrocene of the ferrocenyl ligand as there are no other species initially present. This implies that either the presence of water (or D_2O) or some unknown contaminant can cause the breakdown of some of the ferrocene to release free iron. Ferrocene contains iron(II) whereas this complex contains iron(III), therefore an oxidation step is also required. The iron may be oxidised following release from ferrocene or ferrocene may convert to ferrocenium which then releases iron(III). Other hydrolysis products from the ferrocenyl β-diketonate ligand are likely to be present in solution but these have not been identified.
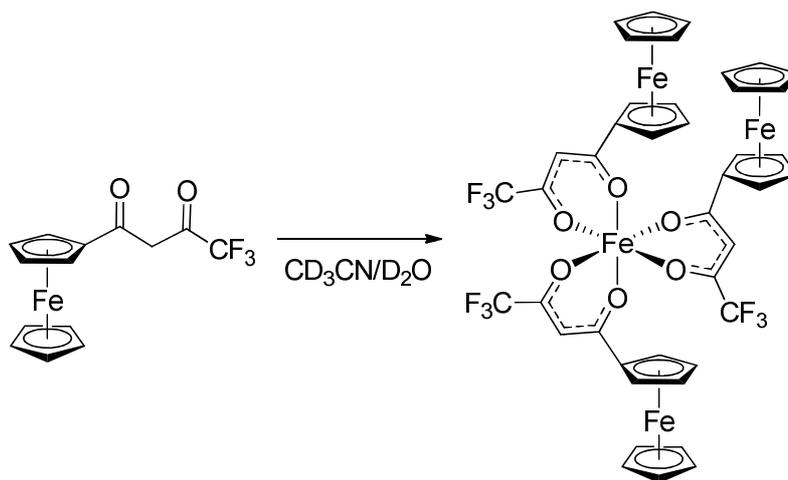
Figure 6.6: Hydrolysis of ferrocenyl β-diketonate ligand component of complex **4.11**

6.2 Hydrophobicity studies

The relative hydrophilicity or lipophilicity (hydrophobicity) of a drug is a key factor in determining its pharmacokinetic profile. Hydrophobicity is determined by calculating the partition coefficient (log P) of a drug. The importance of hydrophobicity is highlighted by the fact that one of the Lipinski rules states that log P should not be greater than 5.[31, 32] Further investigation of "drug-likeness" states that log P should be between -0.4 and 5.6.[33, 34] These intermediate log P values are desired as a drug which is administered orally needs to be sufficiently soluble in both hydrophilic (aqueous) and lipophilic (organic) environments to allow for absorbance across the intestinal lumen into the blood followed by target delivery across a cell membrane.

Hydrophobicity was determined by measuring the octanol-water partition coefficient of selected complexes the using the shake flask-type method, with 1-octanol serving as a model of the lipophilic component. Complexes were dissolved in water-saturated octanol and shaken with an equal volume of octanol-saturated water for 2 hours at 1000 g min^{-1}. The concentration of each complex in the octanol layer, both before and after partitioning, was measured using UV/vis spectroscopy with comparison to previously prepared individual calibration curves. A hydrophobic or lipophilic complex has a positive log P value whereas a hydrophilic complex has a negative log P value.[35, 36]

Hydrophobicity was not investigated for all complexes; only for complexes with activity or partial activity against cancer cell lines (**2.13**, **2.14b**, **2.15b**, **2.16b** and

4.10), their closest analogous inactive complex (**2.14a**, **2.15a**, **2.16a** and **4.11**) and an example of a complex from each of the three series of cobalt picolinamide complexes synthesised (**2.1a**, **2.1b**, **3.2**, **4.3** and **4.13**). The log P values of the investigated complexes are summarised in **Table 6.3** and **Figure 6.7**.

Table 6.3: Log P values of selected cobalt picolinamide complexes

Complex	log P
2.1a	0.10 ± 0.01
2.13	1.54 ± 0.09
2.14a	0.82 ± 0.03
2.15a	1.05 ± 0.03
2.16a	1.57 ± 0.03
2.1b	1.04 ± 0.02
2.14b	0.93 ± 0.01
2.15b	0.72 ± 0.05
2.16b	0.73 ± 0.04
3.1	0.45 ± 0.03
4.3	1.30 ± 0.04
4.10	1.35 ± 0.05
4.11	0.91 ± 0.03
4.13	0.78 ± 0.01
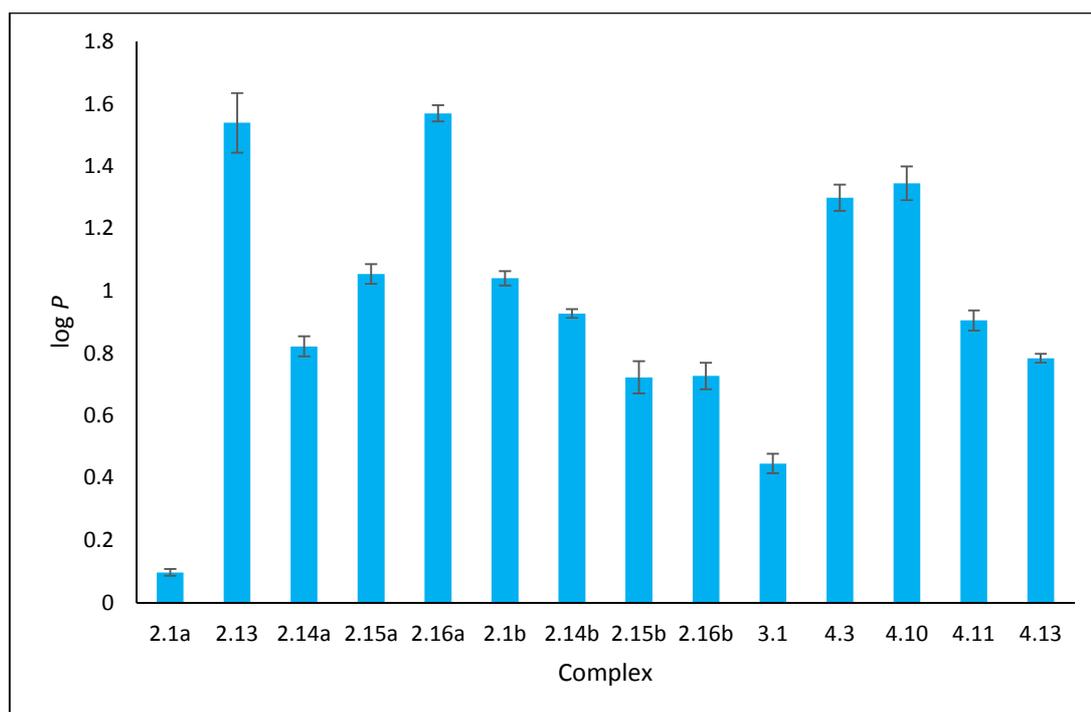
Figure 6.7: Graph of log *P* values of selected cobalt picolinamide complexes

All complexes have positive log *P* values in the range 0.1 - 1.57 and hence are all hydrophobic and within the limits stipulated in the Lipinski rules. The hydrophobic nature of the complexes is unsurprising since the picolinamide ligands contain both pyridyl and aryl rings. In addition, for the cobalt(III) *tris*-picolinamide complexes and cobalt(III) mixed ligand complexes there are three bulky aromatic ligands effectively surrounding the central cobalt ion, thus reducing the hydrophilicity even further. Hydrophobic molecules have a greater potential to cross the lipophilic cell membrane and enter the cell by passive diffusion, implying that these molecules have the potential to enter the cell and that their mode of action is not limited by poor distribution into the cells.[37] However, there are no obvious trends between cytotoxicities and log *P* values suggesting that hydrophobicity is unrelated to anti-cancer potential. Complex **2.13**, one of the two lead complexes, is one of the most hydrophobic complexes (log *P* = 1.54), however complex **2.16a** with a very similar hydrophobicity (log *P* = 1.57) is completely inactive against the cancer cell lines. Complex **2.16b**, the other lead complex, has one of the lower hydrophobicity values (log *P* = 0.73) but again this is very similar to other complexes such as **2.14a** (log *P* = 0.82) which are completely inactive.

Furthermore, there are no obvious trends either within or between the different series of cobalt picolinamide complexes. For example, within the cobalt(III) *tris*-picolinamide series of complexes, for complexes which may adopt either *N,N* or *N,O* coordination, sometimes the *N,N* coordination isomer is more hydrophobic and

sometimes the *N,O* coordination isomer is more hydrophobic. The most interesting hydrophobicity data is for complex **2.1a** (log *P* = 0.10) as this is only slightly hydrophobic with a considerably lower log *P* value than for any of the other complexes. The reason for this is unclear, especially as the analogous complex **2.1b** has an intermediate hydrophobicity (log *P* = 1.04).

6.3 Biomembrane studies

A drug must be able to cross many biological membranes following administration to reach its site of action. Drug distribution, in terms of both concentration and rate, is thus strongly determined by the interactions of a drug with various biomembranes.[38] Therefore, the study of drug-biomembrane interactions is crucial during the preclinical phase to allow compatible drugs to enter clinical trials. Artificial membrane models have been used extensively to study particular aspects of drug-biomembrane interactions due to the highly complex and fragile nature of natural membranes rendering their use labour-intensive.[39, 40] Three types of membrane model are used: monolayers (Langmuir monolayers), vesicle forming bilayers (liposomes) and supported bilayers.[38, 39]

Phospholipid monolayers supported on a mercury on platinum electrode are powerful membrane models with the application of studying the phospholipid monolayer properties in an applied electric field.[41] This sensing electrode is connected to a to a high throughput flow system which uses rapid cyclic voltammetry (RCV) to monitor changes in capacitance current with voltage and allow rapid screening of large numbers of compounds (**Figure 6.8**). The mercury support allows the formation of defect-free, self-healing phospholipid monolayers which allows for the detection of any alterations to monolayer integrity. The monolayers are formed from the phospholipid dioleoyl phosphatidylcholine (DOPC) which acts as the model biomembrane.[40, 42-45]
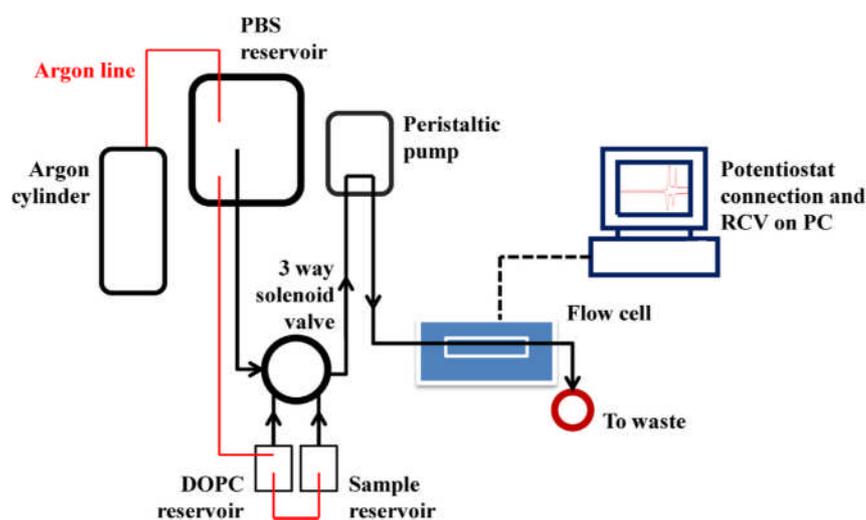
Figure 6.8: Scheme of the model biomembrane system[46]

This unique system, developed by Nelson over the past decade, acts as a model biomembrane to allow the study of drug-biomembrane interactions, ion channel function and the electron transport chain; with the main application as a sensor for biomembrane active molecules.[41, 43, 44] Biomembrane active molecules can interact with the phospholipid monolayer leading to selective membrane damage which is detected electrochemically. This system has been used to detect biomembrane disruption with organic molecules (flavonoids, steroids, polycyclic aromatic hydrocarbons, tricyclic anti-depressants and tricyclic phenothiazines),[46, 47] peptides,[48-51] nanoparticles[52, 53] and ionic liquids.[54] The first use of this system with organometallic compounds was reported recently to detect the membrane disrupting properties of a series of silver(I) non-heterocyclic carbene (NHC) complexes.[55]

The DOPC monolayer undergoes various potential induced phase transitions visualised as sharp peaks in the capacitance current (**Figure 6.9**). In a flow system, such as in this system, two phase transitions are seen as two capacitance current peaks in the RCV plot (**Figure 6.10**). These two peaks correspond to the permeation of electrolyte into the monolayer and the reorganisation of the monolayer into a bilayer.[42, 56]
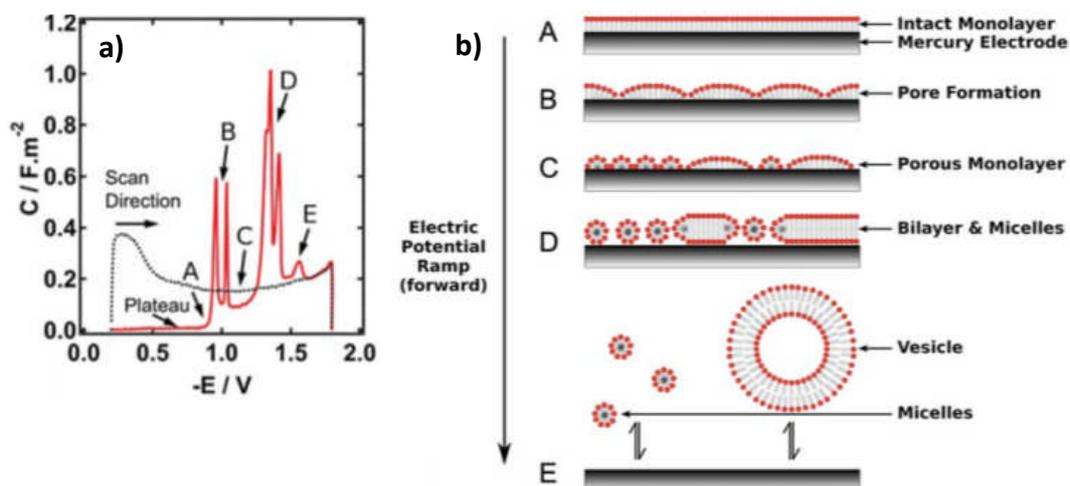
Figure 6.9: a) Typical RCV plot of DOPC monolayer with b) associated phase transitions[56]
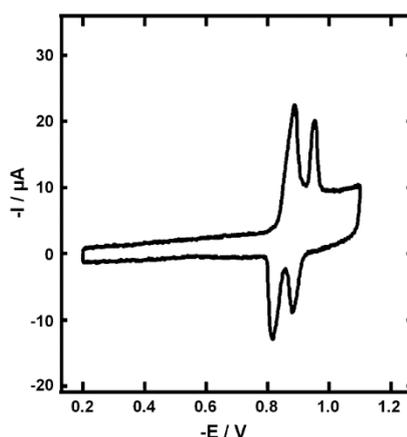
Figure 6.10: Typical RCV plot of DOPC monolayer in a flow cell[42]

Alterations to the organisation or fluidity of the DOPC monolayer alters the phase transitions and thus the characteristic peak shapes, positions and heights in the RCV plot. Reduction in height and broadening of peaks with no change in the baseline current represents adsorption of a molecule onto the monolayer surface. Reduction in the peak heights with an increase in the baseline current represents disruption of the monolayer due to permeation of a molecule.[53]

Complexes were tested for their potential to interact with the artificial biomembrane by Danielle Marriott and Dr. Shahrzad Mohamadi supervised by Professor Andrew Nelson at The University of Leeds. Biomembrane studies of cobalt picolinamide

complexes first requires the deposition of DOPC onto the electrode. A potential excursion from -0.4 to -3.0 V at a scan rate of 100 V s^{-1} is applied, DOPC introduced into the flow cell and the potential excursion altered to -0.4 to -1.2 V. By repetitive cycling, the typical RCV plot are obtained. The complex is then introduced into the flow cell and the RCV plot monitored while cycling the electrode potential from -0.4 to -3.0 V.

Biomembrane interaction was not investigated for all complexes, only cobalt(III) *tris*-picolinamide complexes were investigated as this was the only series of complexes to contain a number of complexes which displayed activity or partial activity against cancer cell lines. Complexes which displayed cytotoxicity (**2.13** and **2.16b**), their closest analogous inactive complex (**2.11** and **2.14b**) and a range of other complexes within this series, encompassing complexes with substituents in the *ortho*, *meta* and *para* positions and complexes with *N,N* and *N,O* coordination (**2.2**, **2.6**, **2.7**, **2.14a**, **2.15a** and **2.1b**). The RCV plots of the interaction of the complexes with the artificial biomembrane are shown in **Figure 6.11**.
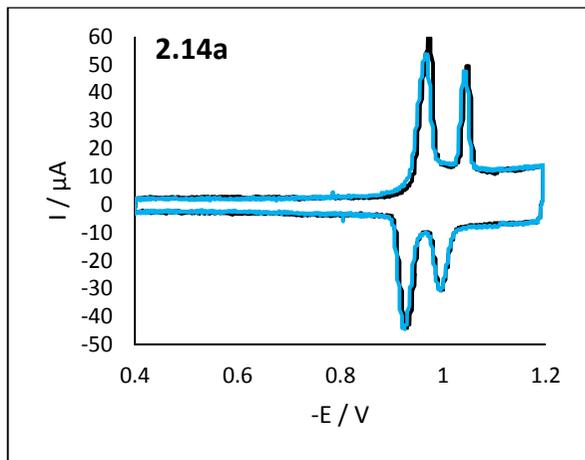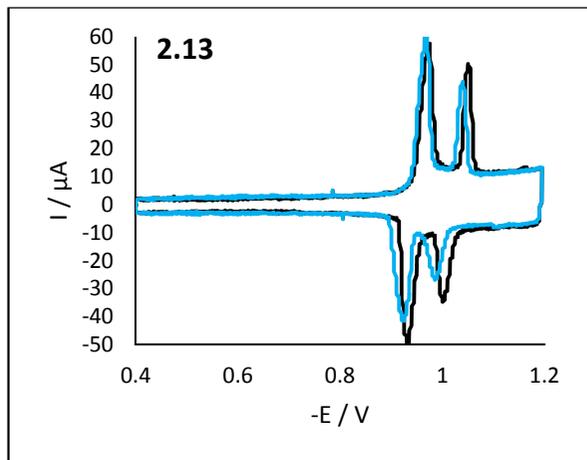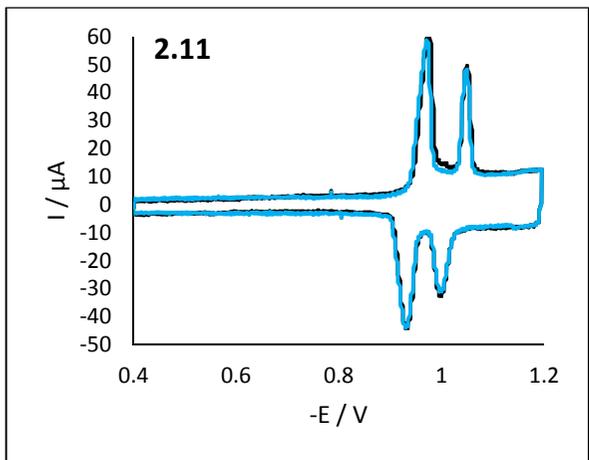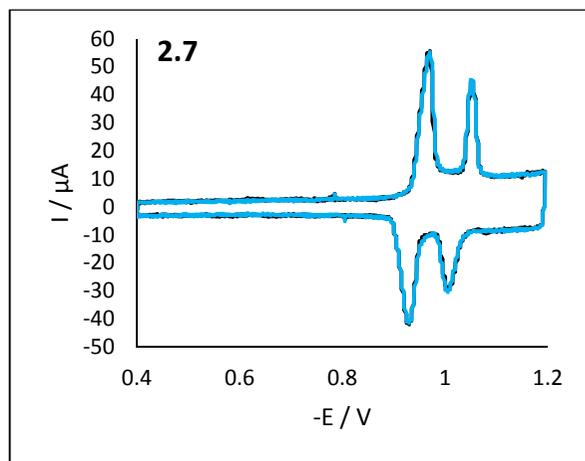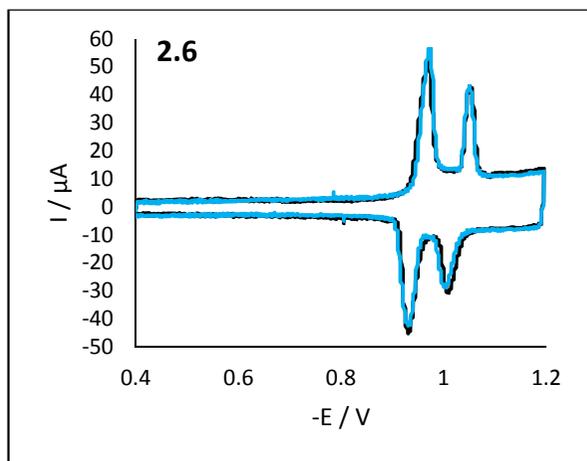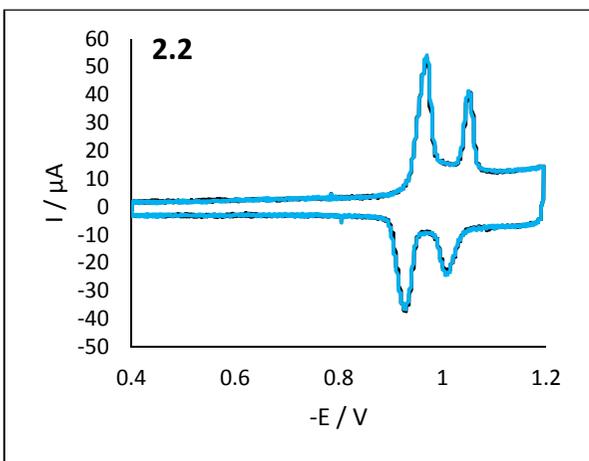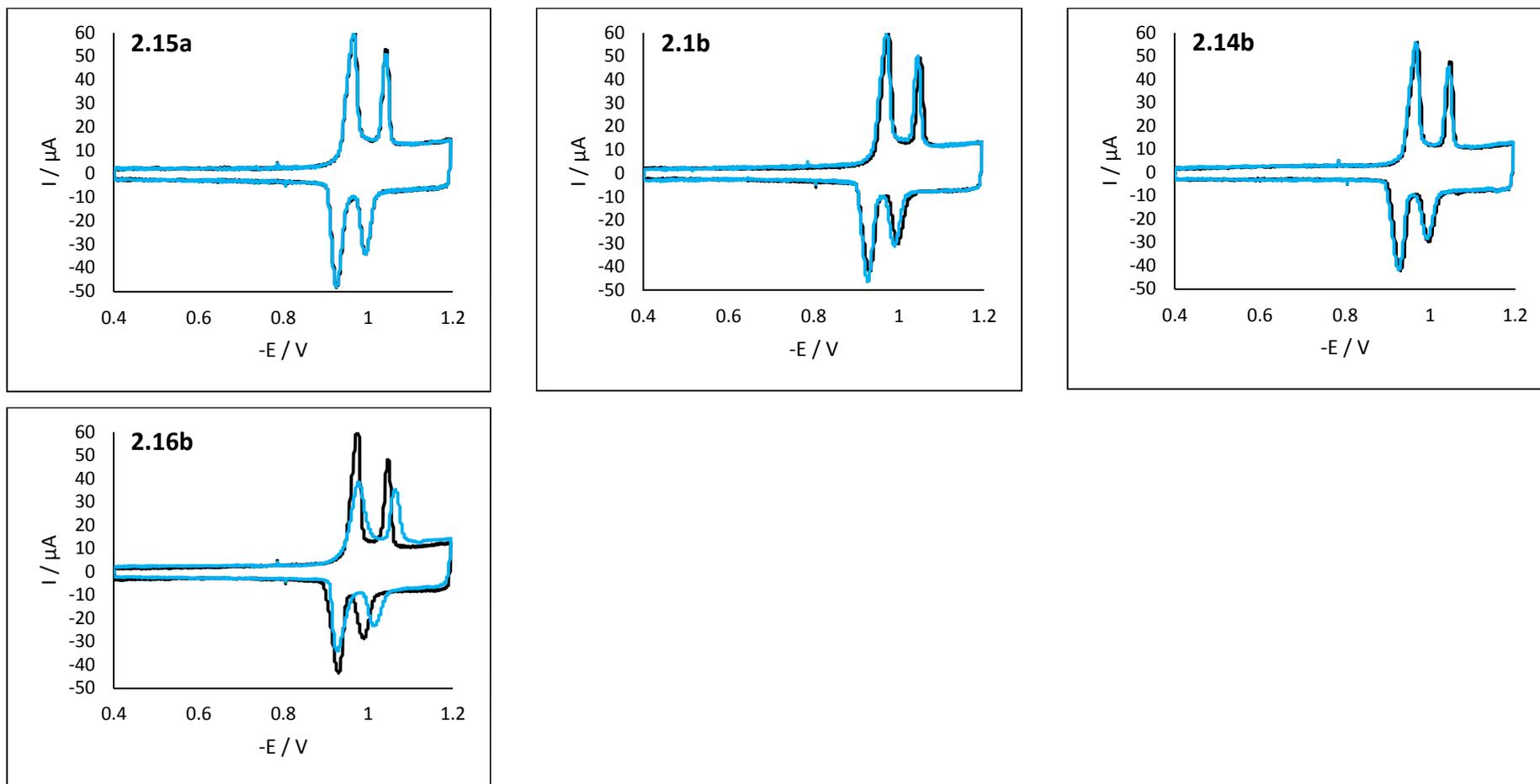
Figure 6.11: RCV plots for the interaction of complexes with the DOPC artificial biomembrane

Key: Black = DOPC membrane in the absence of complex, blue = DOPC membrane in the presence of complex

The only complexes to show distortion of the DOPC membrane RCV plots are **2.13** and **2.16b**. All other complexes show no changes in RCV plots meaning that these complexes are not able to perturb the artificial biomembrane and as such are unable to passively diffuse across the membrane. The implication is that if these complexes enter a cell this can only be achieved through active transport with the aid of membrane channels or transporter proteins. Complexes **2.13** and **2.16b** are able to interact with the biomembrane as visualised by alterations to the RCV plots, namely reduction in the heights and broadening of the peaks with no change in baseline current; these effects are more pronounced for complex **2.16b** compared to **2.13**. These characteristic changes suggest that these complexes are able to adsorb onto the membrane surface rather than being able to permeate the membrane.[53] These results suggest that these complexes also enter a cell through active transport rather than passive diffusion.

The differences in biomembrane interaction between these two complexes and the others tested may be due to the specific shape of these complexes as, for instance complex **2.11** with the 3-(trifluoromethyl) picolinamide ligand shows no membrane interaction whereas complex **2.13** with the 3,5-di(trifluoromethyl) picolinamide ligand does show membrane interaction. Surface charge appears to not have an effect as complexes with electronegative substituents, such as fluoro groups, providing an area of negative surface charge do not perturb the biomembrane despite their potential to interact with the positively charged DOPC head group. In addition, the relative hydrophobicities of the complexes (**Chapter 5.2**) appears to not have an effect as there is no correlation between the hyrdrophobicity data and the ability to interact with the artificial biomembrane.

In addition to being the only complexes tested to display interaction with the biomembrane, complexes **2.13** and **2.16b** are also the lead anti-cancer complexes. This correlation reveals a potential rationale for the activity of these two complexes, perhaps their cytotoxicity is due to a higher degree of passage across the cell membrane leading to improved accumulation within the cell compared to the non-cytotoxic complexes. Complexes **2.13** and **2.16b** are unable to passively diffuse across the biomembrane but are able to adsorb onto the surface, once absorbed onto a cell membrane surface there is the potential for the transfer of these complexes into the cell. This mechanism of internalisation *via* energy-dependent pathways has been previously observed in peptidomimetics and nanoparticles.[57, 58]

6.4 Conclusions

Cobalt(III)-*tris* picolinamide, cobalt(II) *bis*-picolinamide and cobalt(III) mixed ligand complexes with β-diketonate ligands were not observed to undergo hydrolysis. Cobalt(III) mixed ligand complexes with ferrocenyl β-diketonate ligands were observed to hydrolyse through the release of the ferrocenyl β-diketonate ligand resulting in a cobalt *bis*-picolinamide complex. An isolated ferrocenyl β-diketonate ligand has been shown to form an iron(III) *tris*-ferrocenyl β-diketonate complex following hydrolysis, where the iron is hypothesised to originate from the breakdown of ferrocene. The rate of hydrolysis was found to be dependent upon the nature of the substitution of the ferrocenyl β-diketonate ligand with more rapid hydrolysis rates observed with electron withdrawing groups compared to electron donating groups. There was not found to be any correlation between the hydrophobicities of various cobalt picolinamide complexes and cytotoxicity. The interaction of cobalt *tris*-picolinamide complexes with an artificial biomembrane revealed that most complexes were unable to interact with the phospholipid membrane. However, the two lead complexes were able to interact with the biomembrane through adsorption onto the membrane surface. The implication being that these two complexes can undergo more facile entry into a cell through the cell membrane which is the cause of the improved anti-cancer activity.

6.5 References

1.   E. L. Andrade, A. F. Bento, J. Cavalli, S. K. Oliveira, C. S. Freitas, R. Marcon, R. C. Schwanke, J. M. Siqueira and J. B. Calixto, *Brazilian Journal of Medical and Biological Research*, 2016, **49**.

2.   J. P. Hughes, S. Rees, S. B. Kalindjian and K. L. Philpott, *British Journal of Pharmacology*, 2011, **162**, 1239-1249.

3.   ChemoMetec   A/S,   *Two-step   cell   cycle   analysis*, https://chemometec.com/wp-content/uploads/2015/03/994-3001-Two-Step-Cell-Cycle-Assay.pdf, Accessed 02/01/2017, 2017.

4.   B. K. Keppler, C. Friesen, H. G. Moritz, H. Vongerichten and E. Vogel, *Structure and Bonding*, 1991, **78**, 97-127.

5.   B. K. Keppler and M. Hartmann, *Metal-Based Drugs*, 1994, **1**, 145-149.

6.   T. Schilling, K. B. Keppler, M. E. Heim, G. Niebch, H. Dietzfelbinger, J. Rastetter and A. R. Hanauske, *Investigational new drugs*, 1996, **13**, 327-332.

7.   S. P. Gupta, *Chemical Reviews*, 1994, **94**, 1507-1551.

8.   M. M. Hann, A. R. Leach and G. Harper, *Journal of Chemical Information and Computer Sciences*, 2001, **41**, 856-864.

9.   P. Szatkowska-Wandas, M. Koba, G. Smolinski and J. Wandas, *Medicinal Chemistry*, 2016, **12**, 592-600.

10.   R. A. Alderden, M. D. Hall and T. W. Hambley, *Journal of Chemical Education*, 2006, **83**, 728-734.

11.   M. Bouma, B. Nuijen, M. T. Jansen, G. Sava, A. Flaibani, A. Bult and J. H. Beijnen, *International Journal of Pharmaceutics*, 2002, **248**, 239-246.

12.   D. P. Bancroft, C. A. Lepre and S. J. Lippard, *Journal of the American Chemical Society*, 1990, **112**, 6860-6871.

13.   A. Basu and S. Krishnamurthy, *Journal of Nucleic Acids*, 2010, **2010**, Article ID: 201367.

14.   J. K. C. Lau and B. Ensing, *Physical Chemistry Chemical Physics*, 2010, **12**, 10348-10355.

15.   S. H. van Rijt, A. J. Hebden, T. Amaresekera, R. J. Deeth, G. J. Clarkson, S. Parsons, P. C. McGowan and P. J. Sadler, *Journal of Medicinal Chemistry*, 2009, **52**, 7753-7764.

16.   A. M. Basri, R. M. Lord, S. J. Allison, A. Rodriguez-Barzano, S. J. Lucas, F. D. Janeway, H. J. Shepherd, C. M. Pask, R. M. Phillips and P. C. McGowan, *Chem.-Eur. J.*, 2017, **23**, 6341-6356.

17.   A. M. Barrios and S. J. Lippard, *Journal of the American Chemical Society*, 1999, **121**, 11751-11757.

18.   S. Betanzos-Lara, A. Habtemariam, G. J. Clarkson and P. J. Sadler, *European Journal of Inorganic Chemistry*, 2011, 3257-3264.

19.   E. I. Lerner and S. J. Lippard, *Inorganic Chemistry*, 1977, **16**, 1546-1551.

20.   S. Roy, P. U. Maheswari, A. Golobic, B. Kozlevcar and J. Reedijk, *Inorganica Chimica Acta*, 2012, **393**, 239-245.

21.   G. Mestroni, E. Alessio, G. Sava, S. Pacor, M. Coluccia and A. Boccarelli, *Metal-Based Drugs*, 1994, **1**, 41-63.

22.   R. F. Borch and M. E. Pleasants, *Proceedings of the National Academy of Sciences of the United States of America*, 1979, **76**, 6611-6614.

23.   R. C. Dolman, G. B. Deacon and T. W. Hambley, *Journal of Inorganic Biochemistry*, 2002, **88**, 260-267.

24.   A. I. Ivanov, J. Christodoulou, J. A. Parkinson, K. J. Barnham, A. Tucker, J. Woodrow and P. J. Sadler, *Journal of Biological Chemistry*, 1998, **273**, 14721-14730.

25.   J. C. Swarts, T. G. Vosloo, S. J. Cronje, W. C. Du Plessis, C. E. J. Van Rensbur, E. Kreft and J. E. Van Lier, *Anticancer Research*, 2008, **28**, 2781-2784.

26.   M. S. Allison, Unpublished results, 2017.

27.   W. C. du Plessis, T. G. Vosloo and J. C. Swarts, *Journal of the Chemical Society-Dalton Transactions*, 1998, 2507-2514.

28.   K. C. Kemp, E. Fourie, J. Conradie and J. C. Swarts, *Organometallics*, 2008, **27**, 353-362.

29.   P. L. Pauson, *Quarterly Reviews*, 1955, **9**, 391-414.

30.   E. S. Yang, M. S. Chan and A. C. Wahl, *Journal of Physical Chemistry*, 1980, **84**, 3094-3099.

31.   C. Lipinski and A. Hopkins, *Nature*, 2004, **432**, 855-861.

32.   C. A. Lipinski, F. Lombardo, B. W. Dominy and P. J. Feeney, *Advanced Drug Delivery Reviews*, 2001, **46**, 3-26.

33.   A. K. Ghose, V. N. Viswanadhan and J. J. Wendoloski, *Journal of Physical Chemistry A*, 1998, **102**, 3762-3772.

34.   G. Vistoli, A. Pedretti and B. Testa, *Drug Discovery Today*, 2008, **13**, 285-294.

35.   P. D. Leeson and B. Springthorpe, *Nature Reviews Drug Discovery*, 2007, **6**, 881-890.

36.   A. Leo, C. Hansch and D. Elkins, *Chemical Reviews*, 1971, **71**, 525-616.

37.   H. Lodish, A. Berk, S. L. Zipursky, D. Baltimore and J. Darnell, *Molecular cell biology*, Fourth edn., W. H. Freeman, 2000.

38.   R. Pignatello, T. Musumeci, L. Basile, C. Carbone and G. Puglisi, *Journal of Pharmacy and BioAllied Sciences*, 2011, **3**, 4-14.

39.   J. Knobloch, D. K. Suhendro, J. L. Zieleniecki, J. G. Shapter and I. Koper, *Saudi Journal of Biological Sciences*, 2015, **22**, 714-718.

40.   Z. Coldrick, A. Penezic, B. Gasparovic, P. Steenson, J. Merrifield and A. Nelson, *Journal of Applied Electrochemistry*, 2011, **41**, 939-949.

41.   D. Bizzotto and A. Nelson, *Langmuir*, 1998, **14**, 6269-6273.

42.   Z. Coldrick, P. Steenson, P. Millner, M. Davies and A. Nelson, *Electrochimica Acta*, 2009, **54**, 4954-4962.

43.   A. Nelson, *Journal of Electroanalytical Chemistry*, 2007, **601**, 83-93.

44.   A. Nelson, *Current Opinion in Colloid & Interface Science*, 2010, **15**, 455-466.

45.   A. Vakurov, M. Galluzzi, A. Podesta, N. Gamper, A. L. Nelson and S. D. A. Connell, *ACS Nano*, 2014, **8**, 3242-3250.

46.   S. Mohamadi, D. J. Tate, A. Vakurov and A. Nelson, *Analytica Chimica Acta*, 2014, **813**, 83-89.

47.   D. Sanver, B. S. Murray, A. Sadeghpour, M. Rappolt and A. L. Nelson, *Langmuir*, 2016, **32**, 13234-13243.

48.   F. Neville, D. Gidalevitz, G. Kale and A. Nelson, *Bioelectrochemistry*, 2007, **70**, 205-213.

49.   E. Protopapa, S. Maude, A. Aggeli and A. Nelson, *Langmuir*, 2009, **25**, 3289-3296.

50.   E. Protopapa, L. Ringstad, A. Aggeli and A. Nelson, *Electrochimica Acta*, 2010, **55**, 3368-3375.

51.   L. Ringstad, E. Protopapa, B. Lindholm-Sethson, A. Schmidtchen, A. Nelson and M. Malmsten, *Langmuir*, 2008, **24**, 208-216.

52.   N. Ormategui, S. W. Zhang, I. Loinaz, R. Brydson, A. Nelson and A. Vakurov, *Bioelectrochemistry*, 2012, **87**, 211-219.

53.   A. Vakurov, R. Brydson and A. Nelson, *Langmuir*, 2012, **28**, 1246-1255.

54.   M. Galluzzi, S. W. Zhang, S. Mohamadi, A. Vakurov, A. Podesta and A. Nelson, *Langmuir*, 2013, **29**, 6573-6581.

55.   H. A. M. A. Abdelgawad, PhD Thesis, University of Leeds, 2016.

56.   A. V. Brukhno, A. Akinshina, Z. Coldrick, A. Nelson and S. Auer, *Soft Matter*, 2011, **7**, 1006-1017.

57.   X. N. Jing, M. J. Yang, M. R. Kasimova, M. Malmsten, H. Franzyk, L. Jorgensen, C. Foged and H. M. Nielsen, *Biochimica Et Biophysica Acta-Biomembranes*, 2012, **1818**, 2660-2668.

58.   A. Lesniak, A. Salvati, M. J. Santos-Martinez, M. W. Radomski, K. A. Dawson and C. Aberg, *Journal of the American Chemical Society*, 2013, **135**, 1438-1444.

Chapter 7

Conclusions and further work

7. Conclusions and further work

7.1 Conclusions

Picolinamide ligands have proven to be effective ligands for the formation of complexes with a number of different metals.[1-6] One of the interesting features of these ligands is that there are three potential coordination modes to a metal centre: neutral *N,O*-donors or monoanionic *N,O*- or *N,N*-donors (**Figure 7.1**).
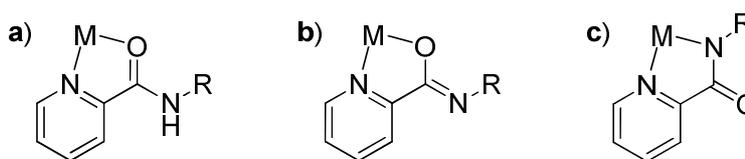
Figure 7.1: Coordination modes for picolinamide ligand a) neutral *N,O*-donor b) monoanionic *N,O*-donor c) monoanionic *N,N*-donor

Previous work within the McGowan group has uncovered the anti-cancer potential of picolinamide complexes of ruthenium, rhodium, iridium and titanium.[7-11] The coordination mode of the picolinamide ligand has also been shown to impact the chemical and biological properties of the complex.[12] The aim of this thesis was the exploration of the anti-cancer potential of a range of cobalt picolinamide complexes.

Three series of cobalt picolinamide complexes have been synthesised and characterised in order to explore their potential biological applications. The first series of complexes are octahedral cobalt(III) *tris*-picolinamide complexes (**Figure 7.2**). Two non-interconverting coordination isomers of these complexes are noted, the major isomer with all three picolinamide ligands displaying *N,N* coordination and the minor isomer with one of these ligands displaying monoanionic *N,O* coordination instead. The minor isomer can only be isolated under certain reaction conditions when the substituent on the aryl ring of the ligand is an electron donating group. The synthetic route for the formation of the major isomer was optimised. However optimisation of the synthetic route for the formation of the minor isomer was unsuccessful.
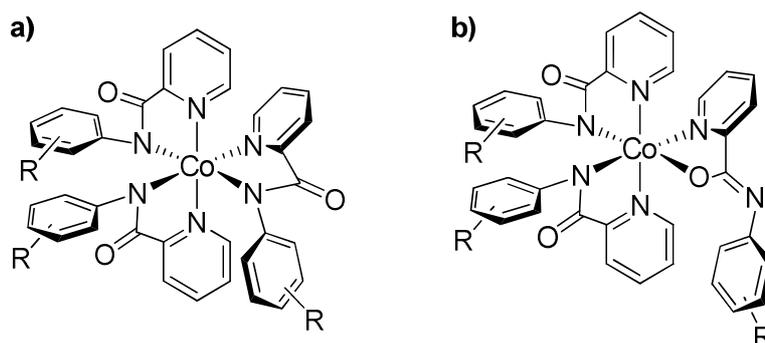
Figure 7.2: Cobalt(III) *tris*-picolinamide complexes with a) *N,N* coordination
b) mixed *N,N* and *N,O* coordination

The second series of complexes are octahedral cobalt(II) *bis*-picolinamide complexes (**Figure 7.3**). These complexes contain two picolinamide ligands with neutral *N,O* coordination and two axial ligand sites which have been occupied with thiocyanate, chloride and aqua ligands. The axial thiocyanate ligands in the cobalt *bis*-picolinamide thiocyanate complexes display either *trans* or *cis* geometry depending upon the substituent on the picolinamide aryl ring. No substituents or substituents in the *meta* position tend to give rise to the *trans* geometry, whereas *ortho* or *para* substituents tend to give rise to complexes with the *cis* geometry. It is hypothesised that the *trans* complex is the thermodynamic product of the synthesis.
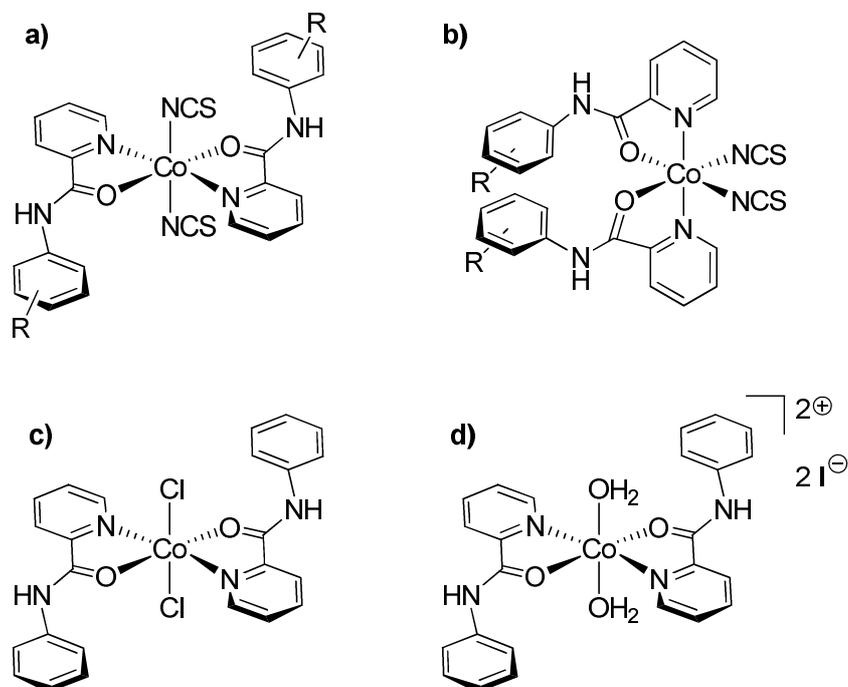
Figure 7.3: Cobalt(II) *bis*-picolinamide complexes with a) *trans* thiocyanate b) *cis* thiocyanate c) *trans* chloride d) *trans* aqua axial ligands

The third series of complexes are octahedral cobalt(III) mixed ligand complexes (**Figure 7.4**). These complexes contain two 3,5-di(trifluoromethyl) picolinamide ligands with *N,N* coordination and one β-diketonate or ferrocenyl β-diketonate ligand. The synthesis of these complexes is controlled stoichiometrically, however a number of side products reduces the experimental yields. The main side products are the cobalt(III) *tris*-picolinamide complex and the cobalt(III) mixed ligand complex with one picolinamide ligand and two β-diketonate ligands.
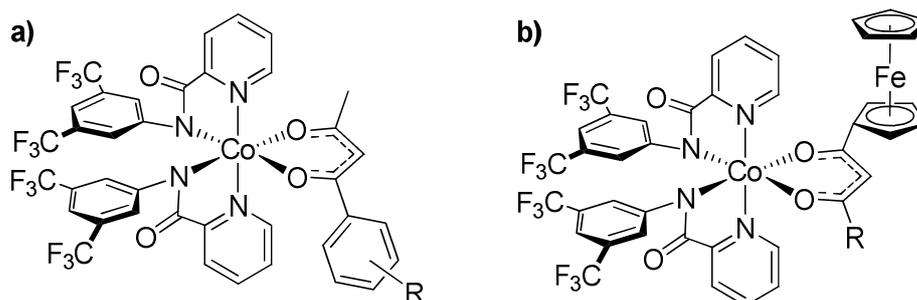
Figure 7.4: Cobalt(III) mixed ligand complexes with a) β-diketonate b) ferrocenyl β-diketonate ligand

The cobalt picolinamide complexes were screened for their anti-cancer, anti-bacterial and anti-fungal properties. Complexes with anti-cancer activity tended to belong to the cobalt(III) *tris*-picolinamide series and complexes with *N,O* coordination were more active than the analogous *N,N* coordination complexes. Cobalt *tris*-picolinamide complexes were tested under hypoxic conditions and found to have lower cytotoxicity compared to when tested under normoxic conditions, implying that these complexes do not undergo activation by reduction. Two lead complexes (**Figure 7.5**) were identified and at physiologically relevant concentrations were found to inhibit cell proliferation through interruption of M phase of the cell cycle. Both complexes were also active against cancer stem cells and were able to inhibit mammosphere formation and viability. The cobalt(II) *bis*-picolinamide thiocyanate complexes were selectively toxic to the fungus *C. albicans*, however activities were only moderate and thus no lead complexes were selected for further study. The thiocyanate ligands were essential for activity; it is theorised that this is due to their ability to undergo ligand substitution leading to the release of free thiocyanate.
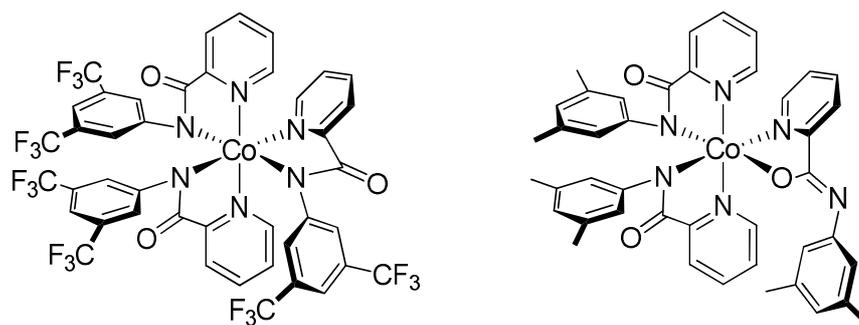
Figure 7.5: Lead anti-cancer cobalt picolinamide complexes

Hydrolysis is often an important mechanism for activation of metal based drugs.[13, 14] The cobalt(III)-*tris* picolinamide, cobalt(II) *bis*-picolinamide and cobalt(III) mixed ligand complexes with β-diketonate ligands did not undergo hydrolysis over a five day period. Cobalt(III) mixed ligand complexes with ferrocenyl β-diketonate ligands underwent hydrolysis, thought to be due to the release of the ferrocenyl β-diketonate ligand, resulting in a cobalt *bis*-picolinamide complex. The rate of hydrolysis was found to be dependent upon the nature of the substitution of the ferrocenyl β-diketonate ligand, with electron withdrawing groups associated with more rapid hydrolysis rates compared to electron donating groups. The hydrophobicities of various cobalt picolinamide complexes was also determined but there was no correlation between log *P* and cytotoxicity. The interaction of cobalt *tris*-picolinamide complexes with an artificial biomembrane was studied. Most complexes were unable to interact with the phospholipid membrane suggesting no role for passive diffusion in the uptake mechanism, whereas the two lead complexes did show an association with the membrane through adsorption onto the membrane surface. The implication is that the two lead complexes may undergo more facile entry into a cell through the cell membrane *via* ATP-dependent mechanisms which is the cause of the improved anti-cancer activity.

7.2 Further work

A variety of substituents on the aryl ring of the picolinamide ligand have been investigated. However a greater number and variety of substituents would provide a greater number of complexes for investigation. This can be performed in a targeted manner following on from the initial results presented in this thesis. For example, a greater number of fluorinated ligands or ligands with substituents in both *meta* positions may be synthesised as both these types of ligand furnished cobalt complexes with anti-cancer activity. Coordinating groups on the picolinamide aryl ring provide the potential to coordinate secondary metal ions, which is thought to be responsible for the anti-cancer and anti-bacterial activity of previous cobalt(III) complexes with bidentate or tridentate picolinamide ligands.[15, 16] The synthesis of similar picolinamide ligands with coordinating groups, e.g. hydroxyl or amine would test this hypothesis in these systems. No investigation into the effect of substitution of the pyridyl ring has been performed, however the high price and limited availability of substituted picolinic acids precludes a detailed investigation *via* the current synthetic route. Alternatively, different heterocycles such as quinaldamides, pyrrolidines or azoles could replace the pyridyl ring.

Another area of the picolinamide ligand which can be modified is the carbonyl group which could be varied as for example an amidine or thioester (**Figure 7.6**) to investigate the impact of changing the inner coordination sphere. Only a brief investigation has been carried out so far with thiopicolinamide ligands which did not result in the formation of any cobalt thiopicolinamide complexes. The methylation of the picolinamide amide group (**Figure 7.6**) would limit the potential binding modes of the ligand to only *N,O* coordination and thus could be used for the formation of further cobalt picolinamide isomers and the investigation into any further differences in biological activity between isomers.
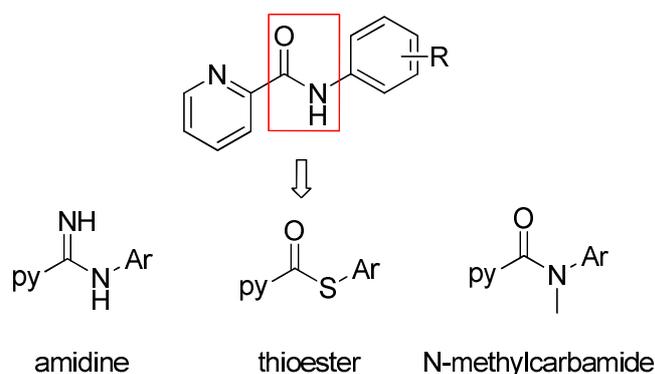
Figure 7.6: Examples of potential picolinamide ligand modifications

Only bidentate picolinamide ligands have been investigated in this thesis; there is also the capacity to investigate both tridentate and tetradentate picolinamide ligands (**Figure 7.7**).
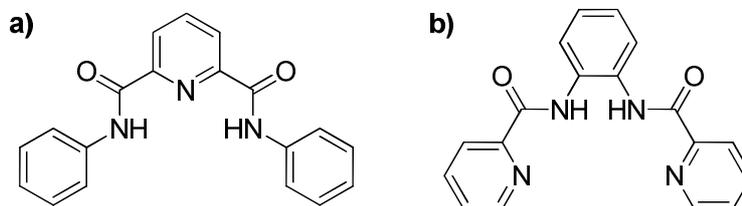
Figure 7.7: General structure of a) tridentate b) tetradentate picolinamide ligands

The cobalt(III) *tris*-picolinamide complexes were only able to form both the *N,N* and *N,O* coordination isomers when the aryl substituent was an electron donating group. However, the number of synthesised ligands containing electron donating groups was limited. The synthesis of further complexes with more electron donating groups, such as *tert*-butyl or amines, would help reveal trends between the strength of electron donation and sterics with the proportion of *N,O* isomer formed and any associated cytotoxicity. The *N,O* coordination isomers showed improved cytotoxicity compared to the analogous *N,N* coordination complexes, however the *N,O* isomer is the minor isomer. The synthesis was not successfully optimised to maximise the formation of this isomer. Previous work with ruthenium(III) picolinamide complexes found that the *N,N* isomer was the thermodynamic product of the synthetic route whereas the *N,O* isomer was the kinetic product and the equilibrium between the formation of these two isomers was temperature and pH dependent.[12] It would be worth investigating whether alterations in time, temperature or pH affect the formation of the *N,O* coordination isomer in this cobalt picolinamide system. In addition, the formation of monocationic cobalt(III) *tris*-picolinamide complexes involving one picolinamide ligand with neutral *N,O* coordination (**Figure 7.8**) with subsequent anti-cancer investigation would allow the full comparison of the different coordination modes in these types of cobalt(III) *tris*-picolinamide complexes.
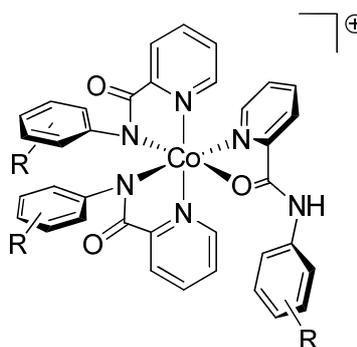
Figure 7.8: General structure of monocationic cobalt(III) *tris*-picolinamide with neutral *N,O* coordination

The cobalt(II) *bis*-picolinamide complexes were only able to form when the aryl ring substituent was an electron donating group. The series could be expanded following the synthesis of a greater number of ligands with a diverse range of electron donating groups attached. Investigations into the synthetic route for complex formation may also allow the formation of complexes with electron withdrawing groups as the aryl substituent, a feat which has yet to be accomplished. It would be interesting to note if the relationship between the aryl substituent position and the *cis/trans* geometry of the cobalt *bis*-picolinamide complex remains the same if electron withdrawing groups are present. The evidence so far suggests that the nature of the cobalt starting material has no effect on the geometry of the final complex, however further syntheses with a greater variety of cobalt salts is needed to confirm this hypothesis. Computational studies into the stability of *cis* and *trans* complexes may reveal the reasons behind the differing geometries adopted and whether one constitutes the thermodynamic product and the other the kinetic product of the reaction.[12]

It is theorised that *trans* and *cis* cobalt(II) bis-picolinamide thiocyanate complexes may have different rates of exchange of the axial thiocyanate ligands. Computational studies can calculate the bond dissociation energies for the cobalt-nitrogen (thiocyanate) bond to discern if there is a difference which may lead to different rates of exchange in solution. Experimental ^{1}H NMR studies with deuterium oxide or labelled groups may also permit investigation of this theory. Adaptations of cobalt(II) *bis*-picolinamide complexes with alternative axial ligands have been synthesised, however there are only a couple of examples of these with the unsubstituted picolinamide ligand. The next step would be the expansion of these series with a greater number of both picolinamide ligands and axial ligands. Alternative axial ligands such as imidazole groups have been synthesised but could not be isolated,

further effort is needed in the investigation into the synthesis and purification of such complexes.

The cobalt(III) mixed ligand complexes synthesised contain two picolinamide ligands with a 3,5-di(trifluoromethyl) group as the aryl substituent and a variable β-diketonate or ferrocenyl β-diketonate ligand. The 3,5-di(trifluoromethyl) picolinamide ligand was selected as the associated cobalt(III) *tris*-picolinamide complex showed activity against various cancerous cell lines. However, the series could be expanded with a number of the other picolinamide ligands synthesised. A greater range of aryl substituents on the β-diketonate ligand can also be investigated, for instance there were no examples of β-diketonate ligands with the aryl substituent in the *ortho* position.

The synthesis of the mixed ligand complex where the β-diketonate ligand was curcumin, was shown to be successful by mass spectrometry but the complex could not be isolated. Further attempts to synthesise and purify this complex are desirable as curcumin is under investigation as a potential anti-cancer drug, thus a cobalt curcumin complex may act as a chaperone to improve the biological properties e.g. bioavailability of curcumin.

Yields of mixed ligand complexes were low due to the formation of a number of side products; optimisation of the synthetic route is required for improvement. One of the side products produced is the cobalt(III) mixed ligand complex with one picolinamide ligand and two β-diketonate ligands (**Figure 7.9**). Synthesis of a series of these compounds would provide an interesting comparison with the series of cobalt(III) picolinamide β-diketonate mixed ligand complexes synthesised so far. In this work only cobalt(III) picolinamide mixed ligand complexes with β-diketonate ligands have been synthesised, but other additional ligands may also be used e.g. β-ketoiminate or amino acid based ligands. The ability to attach diverse ligand types to the mixed ligand complexes would open the opportunity to attach a variety of known drugs to the structure to investigate whether the cobalt(III) picolinamide mixed ligand can act as chaperones.
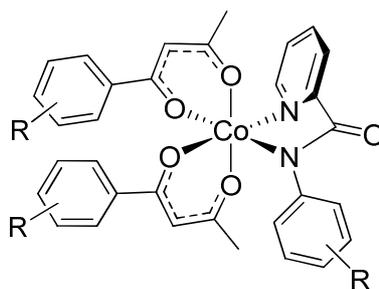
Figure 7.9: Cobalt(III) mixed ligand complex with one picolinamide ligand and two β-diketonate ligands

The anti-cancer activity of the cobalt picolinamide complexes was evaluated against two cancerous cell lines and one non-cancerous cell line. From these investigations, two lead complexes were identified. The evaluation of these two leads against a number of further cancerous cell lines is necessary to examine their potential as anti-cancer drugs against specific cancers. These studies may also uncover underlying targets or mechanisms of action, allowing further cycles of informed drug development. One of the main reason that a potential drug does not progress through clinical trials is toxicity to non-target cells leading to adverse drug reactions.[17] Toxicity to hepatocytes is frequently seen as the liver is the main organ responsible for the metabolism of drugs. Investigation into the potential toxicity of the two lead complexes to hepatocytes can be investigated with various *in vitro* studies, for example using cultured hepatocytes or liver slices.[18]

The investigation into the performance of complexes in a hypoxic environment was only performed with the cobalt(III) *tris*-picolinamide complexes, the behaviour of the cobalt(III) mixed ligand complexes under hypoxia should also be investigated. Hypoxic studies of cobalt(III) *tris*-picolinamide complexes suggests that these complexes may undergo bioreduction to inactive cobalt(II) *bis*-picolinamide complexes. The reduction potential of this Co(III)/Co(II) couple could be determined for the cobalt(III) *tris*-picolinamide complexes using cyclic voltammetry to ascertain whether this reduction is biologically feasible.

At physiologically relevant concentrations, the two lead complexes were found to induce cell cycle arrest at M phase. However, due to the low proportion of cells in M phase at any given time point any changes in population are small. Therefore, further investigation of these complexes at higher concentrations and shorter exposure times would be beneficial as these conditions should reduce the amount of natural cell death and reduce the number of aggregated cells, thus more

accurately presenting the effects of the complexes on the cell cycle. Most drugs which act at M phase of the cell cycle inhibit the polymerisation of tubulin, which is essential for spindle formation; the ability of the two lead complexes to bind tubulin and inhibit tubulin polymerisation should be investigated. Further investigations to uncover the mechanism of action of these lead complexes are essential, for example investigations into any anti-metastatic properties, the ability to bind to DNA, induction of changes in protein expression or interaction with various intracellular targets.

The cobalt(II) *bis*-picolinamide thiocyanate complexes were shown to have moderate anti-fungal activity. Previous cobalt complexes in the literature with anti-fungal properties have shown decreased activity compared to their copper analogues.[19, 20] Therefore the synthesis of the analogous copper(II) *bis*-picolinamide thiocyanate complexes and evaluation of their anti-fungal properties would be of interest.

Investigation into the chemical properties of a range of cobalt picolinamide complexes found that cobalt(III) *tris*-picolinamide complexes and cobalt(III) mixed ligand complexes with β-diketonate ligands did not undergo hydrolysis. However, these investigations were performed with 20 % water, due to the lack of solubility of some complexes at higher percentages of water. For complexes which are soluble at higher concentrations of water it should be confirmed that the behaviour of the complexes does not alter. Cobalt(III) mixed ligand complexes with ferrocenyl β-diketonate ligands underwent hydrolysis with 20 % water. If these complexes are sufficiently soluble, this behaviour should be confirmed with different concentrations of water, to determine if a particular threshold level of water is required or if the behaviour or resultant hydrolytic products differ with changes in concentration. For complexes which undergo hydrolysis, the rates of hydrolysis and the amount of hydrolysis should be quantified as previous work within the McGowan group with ruthenium(III) *bis*-picolinamide complexes has shown a correlation between the amount of hydrolysis and cytotoxicity. The current rates of hydrolysis appear to depend upon the nature of the ligand substituent, with more electron withdrawing groups leading to more rapid hydrolysis compared to electron donating groups. This hypothesis should be confirmed by monitoring the hydrolysis of all of the cobalt(III) mixed ligand complexes with ferrocenyl β-diketonate ligands. The determination of the hydrolytic products of the complexes would be informative, at present only one product from the hydrolysis of a ferrocenyl β-diketonate ligand as opposed to the complex itself has been identified through X-ray crystallography.

Through artificial biomembrane studies, it was found that the two lead complexes could interact with the membrane whereas the inactive complexes did not show any interaction. This implies that the uptake of the lead complexes into the cell is improved compared to the inactive complexes. This result with the artificial biomembrane should be confirmed with *in vitro* studies using inductively coupled plasma mass spectrometry (ICP-MS) to determine the intracellular cobalt concentration (and distribution) before and after incubation with the two lead complexes. Only cobalt(III) *tris*-picolinamide complexes were investigated, therefore investigation of the other series of cobalt picolinamide complexes would be instructive to determine if these are inactive due to poor membrane interaction.

Cobalt(III) *tris*-picolinamide complexes have previously been noted to be active and selective catalysts for the oxidation of ethylbenzene to acetophenone with oxygen as the oxidant.[21] No catalytic studies have been performed on any of the cobalt picolinamide complexes in this work. The evaluation of all the cobalt picolinamide complexes as potential catalysts for the oxidation of ethylbenzene and other organic compounds is highly desirable.

7.3 References

1.   R. L. Chapman, F. S. Stephens and R. S. Vagg, *Inorganica Chimica Acta-Articles*, 1981, **52**, 161-168.

2.   G. Condorelli, A. Seminara and A. Musumeci, *Journal of Inorganic & Nuclear Chemistry*, 1974, **36**, 3763-3766.

3.   K. Nonoyama, *Journal of Inorganic & Nuclear Chemistry*, 1975, **37**, 2449-2453.

4.   M. Ray, R. Mukherjee, J. F. Richardson and R. M. Buchanan, *Journal of the Chemical Society-Dalton Transactions*, 1993, 2451-2457.

5.   G. V. Tsintsadze, R. A. Kiguradze, A. N. Shnulin and K. S. Mamedov, *Journal of Structural Chemistry*, 1986, **27**, 272-278.

6.   Z. Q. Xu, L. K. Thompson, D. A. Black, C. Ralph, D. O. Miller, M. A. Leech and J. A. K. Howard, *Journal of the Chemical Society-Dalton Transactions*, 2001, 2042-2048.

7.   Z. Almodares, S. J. Lucas, B. D. Crossley, A. M. Basri, C. M. Pask, A. J. Hebden, R. M. Phillips and P. C. McGowan, *Inorganic Chemistry*, 2014, **53**, 727-736.

8.   A. M. Basri, R. M. Lord, S. J. Allison, A. Rodriguez-Barzano, S. J. Lucas, F. D. Janeway, H. J. Shepherd, C. M. Pask, R. M. Phillips and P. C. McGowan, *Chem.-Eur. J.*, 2017, **23**, 6341-6356.

9.   K. D. Camm, A. El-Sokkary, A. L. Gott, P. G. Stockley, T. Belyaeva and P. C. McGowan, *Dalton Transactions*, 2009, 10914-10925.

10.   S. Lord, University of Leeds, 2007.

11.   S. J. Lucas, R. M. Lord, R. L. Wilson, R. M. Phillips, V. Sridharan and P. C. McGowan, *Dalton Transactions*, 2012, **41**, 13800-13802.

12.   S. H. van Rijt, A. J. Hebden, T. Amaresekera, R. J. Deeth, G. J. Clarkson, S. Parsons, P. C. McGowan and P. J. Sadler, *Journal of Medicinal Chemistry*, 2009, **52**, 7753-7764.

13.   R. A. Alderden, M. D. Hall and T. W. Hambley, *Journal of Chemical Education*, 2006, **83**, 728-734.

14.   M. Bouma, B. Nuijen, M. T. Jansen, G. Sava, A. Flaibani, A. Bult and J. H. Beijnen, *International Journal of Pharmaceutics*, 2002, **248**, 239-246.

15.   N. K. Kaushik, A. Mishra, A. Ali, J. S. Adhikari, A. K. Verma and R. Gupta, *Journal of Biological Inorganic Chemistry*, 2012, **17**, 1217-1230.

16. A. Mishra, N. K. Kaushik, A. K. Verma and R. Gupta, *European Journal of Medicinal Chemistry*, 2008, **43**, 2189-2196.

17. J. P. Hughes, S. Rees, S. B. Kalindjian and K. L. Philpott, *British Journal of Pharmacology*, 2011, **162**, 1239-1249.

18. V. Y. Soldatow, E. L. LeCluyse, L. G. Griffith and I. Rusyn, *Toxicology Research*, 2013, **2**, 23-39.

19. N. V. Loginova, T. V. Koval'chuk, G. I. Polozov, N. P. Osipovich, P. G. Rytik, Kucherov, II, A. A. Chernyavskaya, V. L. Sorokin and O. I. Shadyro, *European Journal of Medicinal Chemistry*, 2008, **43**, 1536-1542.

20. P. V. Rao, S. Ammanni and S. Kalidasu, *E-Journal of Chemistry*, 2011, **8**, 470-478.

21. J. Y. Qi, H. X. Ma, X. J. Li, Z. Y. Zhou, M. C. K. Choi, A. S. C. Chan and Q. Y. Yang, *Chemical Communications*, 2003, 1294-1295.

Chapter 8

Experimental data and protocols

8. Experimental section

8.1 General experimental procedures

All ligands and complexes were synthesised under aerobic conditions. All chemicals were supplied by Sigma-Aldrich Chemical Co., Acros Organics, Alfa Aesar and BOC gases. Deuterated NMR solvents were purchased from Sigma-Aldrich Chemical Co. or Acros Organics.

8.2 Instrumentation

All NMR spectra were acquired using a Bruker Avance 300, 400 or 500 MHz spectrometer. Chemical shifts (δ) are expressed in parts per million (ppm) and referred to the solvent signal, used as an internal reference. Mass spectra were recorded at the University of Leeds Mass Spectrometry Service on a Bruker Daltonics MicroTOF instrument with electrospray ionisation (ESI) and a photodiode array analyser. Microanalyses were acquired either by Ms. Tanya Marinko-Covell at the University of Leeds Microanalytical Service using a Carlo Erba 1108 Elemental Analyser or by Mr. Stephen Boyer at the London Metropolitan University Elemental Analysis Service. Infrared spectra were acquired on a Perkin-Elmer SpectrumOne FT-IR spectrometer. UV/vis absorption spectra were acquired on a Cary Series UV-Vis spectrophotometer using 1 cm path length quartz cuvettes. Magnetic susceptibility measurements were performed by Dr. Rafal Kulmaczewski at the University of Leeds on a Quantum Design VSM SQUID magnetometer in an applied field of 500 G.

8.3 X-Ray crystallography

Single crystal X-ray diffraction data were collected by the author or Dr. Christopher Pask using an Agilent (Rigaku) SuperNova X-ray diffractometer fitted with an Atlas area detector using mirror monochromated Mo-K$_\alpha$ (λ = 0.71073 Å) or Cu-K$_\alpha$ (λ = 1.54184 Å) radiation. The crystal was cooled to 120 K using an Oxford Cryosystem low temperature device.[1] The full dataset was collected and the images processed using CrysAlisPro program.[2] Structure solution by direct methods was achieved through the use of SHELXS86,[3] SHELXL-2014[4] or SHELXT,[5] and the structural model was refined by full matrix least squares on F^2 using SHELX97[6] interfaced through the program Olex2.[7] Molecular graphics were plotted, editing of CIFs and construction

of tables of bond lengths and angles were achieved using Olex2. Unless otherwise stated, hydrogen atoms were placed using idealised geometric positions (with free rotation for methyl groups), allowed to move in a "riding model" along with the atoms to which they were attached, and refined isotropically. The SQUEEZE routine of Platon was used to refine structures where diffuse electron density could not be adequately modelled as solvent of crystallisation.[8]

8.4 X-Ray powder diffraction

X-ray powder diffraction data were collected by the author or Dr. Christopher Pask using a Bruker AXS D2Phaser diffractometer. Data collection was carried out at room temperature using Cu-K$_\alpha$ (λ = 1.54184 Å) radiation. Diffraction patterns were recorded in step-scan mode with a step size of 0.2° from 5° to 50° (5 s per step) using a 0.1 mm divergent slit. Experimental data was processed using Diffrac.Suite Eva[9] and Chekcell.[10] Mercury was used to simulate the powder diffraction pattern from the single crystal structures.[11]

8.5 Syntheses of N-aryl picolinamide ligands

Ligands **1.1** - **1.21** have been previously reported in the literature;[12-17] only the characterisation of novel ligand **1.22** is reported in detail. Ligands **1.2** - **1.8** and **1.10** were synthesised and characterised by Dr. Carlo Sambiagio, Dr. Aida Basri or Dr. Christopher Pask (University of Leeds).

8.5.1 General synthetic procedure for N-aryl picolinamide ligands

Ligands **1.1** - **1.10** and **1.12** - **1.22** were synthesised *via* a literature method.[18] The required aniline (1 equivalent) was added to a solution of pyridine-2-carboxylic acid (1 equivalent) in pyridine and warmed to 50 °C for 15 minutes. To this mixture, triphenylphosphite (1 equivalent) was added and heated at 110 °C for 18 hours. After cooling to room temperature, addition of distilled water precipitated out the crude product (for ligands **1.2** - **1.10** and **1.12** - **1.14**) which was isolated by filtration, washed with distilled water and purified by recrystallisation from hot methanol. For ligands **1.1** and **1.15** - **1.22**, addition of distilled water instead yielded an oil, dichloromethane was added to the mixture and the organic layer separated from the aqueous layer. The product in the organic layer was extracted three times with 1:1 (v/v) aqueous hydrochloric acid. To neutralise the acid extract, sodium

bicarbonate was added until pH 7 was reached. The crude product was isolated by filtration, washed with distilled water and purified by recrystallisation from hot methanol.

8.5.2 General synthetic procedure for ligand 1.11

Ligand **1.11** was synthesised *via* an alternative literature method.[19] Pyridine-2-carboxylic acid (1 equivalent), the required aniline (1 equivalent) and triethylamine (2 equivalents) were dissolved in dichloromethane and stirred at 0 °C. To this mixture, phosphorus(V) oxychloride (2 equivalents) was added dropwise and stirred at 0 °C for 15 minutes. The mixture was gradually warmed to room temperature while stirring for 2 hours. Excess phosphorus(V) oxychloride was quenched by slow addition of distilled water, then the product in the organic layer was extracted with dichloromethane. Organic fractions were combined and the evaporated to give the crude product which was purified by recrystallisation from hot pentane.

8.5.3 Synthesis of $C_{20}H_{14}N_2O$ (1.22)

Brown solid (0.962 g, 3.225 mmol, 64 %). Recrystallisation by vapour diffusion (chloroform/pentane) at room temperature yielded yellow crystals suitable for single crystal X-ray diffraction.
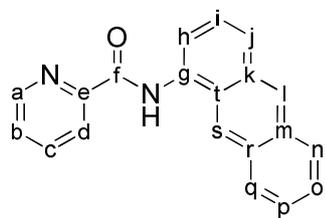
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.0 K) δ 10.93 (br. s, 1H, NH), 8.81 (d, 1H, H$_a$, ^3J (^{1}H-^{1}H) = 4.6 Hz), 8.64 (s, 1H, H$_l$), 8.50 (s, 1H, H$_s$), 8.43 (d, 1H, H$_h$, ^3J (^{1}H-^{1}H) = 2.5 Hz), 8.42 (d, 1H, H$_d$, ^3J (^{1}H-^{1}H) = 3.2 Hz), 8.13 (m, 1H, H$_n$ or H$_q$), 8.04 (m, 1H, H$_n$ or H$_q$), 8.00 (td, 1H, H$_c$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.6 Hz), 7.89 (d, 1H, H$_j$, ^3J (^{1}H-^{1}H) = 8.5 Hz), 7.59 (ddd, 1H, H$_b$, ^3J (^{1}H-^{1}H) = 7.7 Hz, ^3J (^{1}H-^{1}H) = 4.9 Hz and ^4J (^{1}H-^{1}H) = 1.1 Hz), 7.52 (m, 3H, H$_i$, H$_o$ and H$_p$) **^{13}C{^{1}H} NMR** (CDCl$_3$, 125.9 MHz, 299.3 K) δ 162.4 (quaternary C-O, C$_f$), 150.2 (quaternary aniline C, C$_e$), 148.2 (aniline CH, C$_a$), 137.8 (aniline CH, C$_c$), 132.2 (quaternary aromatic C, C$_k$), 132.1 (quaternary aromatic C, C$_t$), 131.6 (aromatic CH, C$_o$ and C$_p$), 128.5 (aromatic CH, C$_n$ or C$_q$), 128.0 (aromatic CH, C$_n$ or C$_q$), 127.3 (aromatic CH, C$_s$), 126.6 (aniline CH, C$_b$), 125.8 (quaternary aromatic C, C$_m$), 125.7 (quaternary aromatic C, C$_r$), 125.5 (quaternary aromatic C, C$_g$), 125.4 (aromatic CH, C$_j$), 125.3 (aromatic CH, C$_i$), 122.6 (aniline CH, C$_d$), 119.1 (aromatic CH, C$_l$), 117.5 (aromatic CH, C$_h$) **Analysis calculated** C 80.52, H 4.73, N 9.39 % **Analysis found** C 80.20, H 4.60, N 9.20 % **ES MS (+)** (CH$_3$OH) *m/z* 299.12 [MH$^+$] **IR** (cm^{-1}) ν 3352 (m, NH), 3048 (w, Ar CH), 1683 (m, CO), 1540 (m), 1516 (s), 1460 (m), 1400 (m), 1347 (m), 1210 (w), 1136 (w), 1014 (w), 843 (s), 728 (s), 632 (s), 531 (s)

8.6 Syntheses of β-diketonate ligands

β-Diketonate ligands have been previously reported in the literature[20-22] and thus the characterisation data is not reported here. Aside from the 4-(trifluoromethyl) β-diketonate ligand, all ligands were synthesised and characterised by Dr. Felix Janeway or Dr. Andrew Hebden (University of Leeds).

8.6.1 General synthetic procedure for β-diketonate ligands

Ligands were synthesised *via* a literature method.[23] Sodium ethoxide (1 equivalent) was added to a solution of the required acetophenone (1 equivalent) in ethyl acetate and heated at 80 °C for 4 hours. After cooling to room temperature, ice cold sulfuric acid was added dropwise until the mixture was just acidic. The crude product was extracted three times into diethyl ether, dried over magnesium sulfate, filtered and the solvent removed under reduced pressure to yield the pure product.

8.7 Syntheses of cobalt(III) *tris*-picolinamide complexes (*N,N* coordination)

The required picolinamide ligand (3 equivalents) in ethanol (10 mL) was added to a solution of cobalt(II) chloride hexahydrate (1 equivalent) in distilled water (10 mL). Triethylamine (3 equivalents) was added and the mixture was heated at 85 °C for 2 hours to give a brown solution. The solvent was removed under reduced pressure and the product purified by column chromatography (methanol/diethyl ether). Product was dissolved in dichloromethane or chloroform, filtered through a sinter and the pure product was precipitated with the addition of hexane.

8.7.1 Synthesis of CoC$_{36}$H$_{27}$N$_6$O$_3$ (2.1a)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a red solid (0.225 g, 0.346 mmol, 82 %). Recrystallisation by vapour diffusion (acetonitrile/diethyl ether) at 5 °C yielded red crystals suitable for single crystal X-ray diffraction.
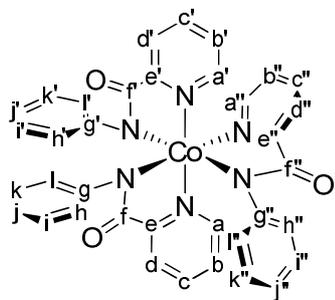
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.2 K) δ 9.17 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.59 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.7 Hz), 8.37 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.12 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.4 Hz), 7.97 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 7.78 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.1 Hz), 7.73 (m, 2H, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ and H$_{d''}$), 7.66 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.3 Hz), 7.26 (m, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 7.17 (m, 2H, H$_a$, H$_{a'}$ or H$_{a''}$ and H$_b$, H$_{b'}$ or H$_{b''}$), 7.14 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 6.95 (m, 6H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_j$, H$_{j'}$ or H$_{j''}$), 6.81 (t, 1H, H$_j$, H$_{j'}$ or H$_{j''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 6.71 (t, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz), 6.49 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.12 (br. s, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.1 K) δ 170.4 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.5 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 168.9 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 158.5 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 157.2 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 156.7 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 149.7 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 149.0 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.4 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 145.9 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 144.3 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 143.7 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 140.5 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.5 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.4 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 129.3 (aromatic C̲H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$, and C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 128.8 (aromatic C̲H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 128.1 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$ and C$_b$, C$_{b'}$ or C$_{b''}$), 127.7 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.7 (aromatic C̲H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$, and C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 126.6 (aromatic C̲H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 125.9 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.8 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 125.5 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$), 125.1 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$), 124.8 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$) **Analysis calculated** (+ 2 H$_2$O) C 62.97, H 4.55, N 12.24 % **Analysis found** C 63.40, H 4.00, N 11.90 % **ES MS (+)** (CH$_3$OH) m/z 651.16 [MH$^+$] **IR** (cm^{-1}) ν 3051 (w, ArCH), 1620 (s, CO), 1585 (s, CO), 1567 (s, CO), 1487 (m), 1364 (m), 1289 (w), 1145 (w), 759 (m), 685 (m), 503 (m)

8.7.2 Synthesis of CoC$_{36}$H$_{24}$N$_6$O$_3$F$_3$ (2.2)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a brown solid (0.220 g, 0.313 mmol, 75 %). Recrystallisation by slow evaporation (ethanol/water) at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
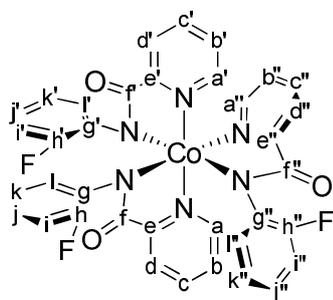
^{1}**H NMR** (CDCl$_3$, 500.23 MHz, 300.1 K) δ 9.24 (br. s, 1H, H$_a$, H$_{a'}$ or H$_{a''}$), 8.78 (t, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 6.2 Hz), 8.31 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.07 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.2 Hz), 8.02 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 7.84 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz), 7.75 (m, 2H, H$_c$, H$_{c'}$ or H$_{c''}$and H$_d$, H$_{d'}$ or H$_{d''}$), 7.66 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.4 Hz, ^3J (^{1}H-^{1}H) = 5.9 Hz and ^4J (^{1}H-^{1}H) = 1.4 Hz), 7.58 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.2 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.3 Hz), 7.50 (br. s, 1H, H$_a$, H$_{a'}$ or H$_{a''}$), 7.23 (m, 3H, H$_i$, H$_{i'}$ or H$_{i''}$, H$_k$, H$_{k'}$ or H$_{k''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 7.12 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.3 Hz), 6.95 (m, 2H, H$_k$, H$_{k'}$ or H$_{k''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 6.86 (m, 2H, H$_i$, H$_{i'}$ or H$_{i''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 6.50 (m, 3H, H$_i$, H$_{i'}$ or H$_{i''}$, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 5.42 (t, 1H, H$_j$, H$_{j'}$ or H$_{j''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz), 5.27 (t, 1H, H$_j$, H$_{j'}$ or H$_{j''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz) ^{13}**C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.1 K) δ 169.9 (quaternary C-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.5 (quaternary C-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.1 (quaternary C-O, C$_f$, C$_{f'}$ or C$_{f''}$), 158.0 (quaternary aniline C, C$_e$, C$_{e'}$ and C$_{e''}$), 150.0 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$), 149.9 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$), 149.6 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$), 151.1 (quaternary aromatic C, C$_h$, C$_{h'}$ or C$_{h''}$), 150.9 (quaternary aromatic C, C$_h$, C$_{h'}$ or C$_{h''}$ and C$_h$, C$_{h'}$ or C$_{h''}$), 140.4 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 139.8 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 139.3 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 133.2 (quaternary aromatic C, C$_g$, C$_{g'}$ or C$_{g''}$), 131.8 (quaternary aromatic C, C$_g$, C$_{g'}$ or C$_{g''}$), 130.9 (quaternary aromatic C, C$_g$, C$_{g'}$ or C$_{g''}$), 127.8 (aromatic CH, C$_j$, C$_{j'}$ or C$_{j''}$), 127.3 (aromatic CH, C$_j$, C$_{j'}$ or C$_{j''}$ and C$_j$, C$_{j'}$ or C$_{j''}$), 127.2 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$), 126.9 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$), 126.8 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$), 126.3 (aniline CH, C$_d$, C$_{d'}$ or C$_{d''}$), 126.0 (aniline CH, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 125.3 (aromatic CH, C$_k$, C$_{k'}$ or C$_{k''}$), 125.2 (aromatic CH, C$_k$, C$_{k'}$ or C$_{k''}$ and C$_k$, C$_{k'}$ or C$_{k''}$), 124.9 (aromatic CH, C$_l$, C$_{l'}$ or C$_{l''}$), 123.8 (aromatic CH, C$_l$, C$_{l'}$ or C$_{l''}$ and C$_l$, C$_{l'}$ or C$_{l''}$), 115.7 (aromatic CH, C$_i$, C$_{i'}$ or C$_{i''}$), 115.5 (aromatic CH, C$_i$, C$_{i'}$ or C$_{i''}$), 115.3 (aromatic CH, C$_i$, C$_{i'}$ or C$_{i''}$) **Analysis calculated** (+ 1 H$_2$O) C 59.84, H 3.63, N 11.63 % **Analysis found** C 59.70, H 3.40, N 11.48 % **ES MS (+)** (CH$_3$OH) *m/z* 705.13 [MH$^+$] **IR** (cm^{-1}) ν 3080 (w, ArCH), 1625 (m, CO), 1590 (m, CO), 1489 (m), 1362 (m), 1096 (w), 762 (s), 687 (m), 508 (m)

8.7.3 Synthesis of CoC$_{36}$H$_{24}$N$_6$O$_3$F$_3$ (2.3)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a brown solid (0.192 g, 0.272 mmol, 65 %). Recrystallisation by vapour diffusion (methanol/diethyl ether) at 5 °C yielded red crystals suitable for single crystal X-ray diffraction.
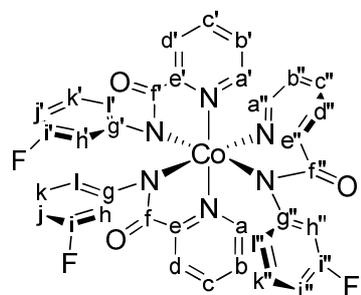
^{1}H NMR (CDCl$_3$, 500.23 MHz, 299.9 K) δ 9.03 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 8.55 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 8.38 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 8.16 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.2 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.99 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 7.86 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.2 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.78 (m, 2H, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.69 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 6.8 Hz, ^3J (^{1}H-^{1}H) = 5.5 Hz and ^4J (^{1}H-^{1}H) = 1.1 Hz), 7.24 (m, 5H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_h$, H$_{h'}$ or H$_{h''}$ and H$_j$, H$_{j'}$ or H$_{j''}$), 6.98 (m, 3H, H$_h$, H$_{h'}$ or H$_{h''}$, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 6.69 (m, 2H, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 6.54 (t, 1H, H$_l$, H$_{l'}$ or H$_{l''}$, ^3J (^{1}H-^{1}H) = 8.0 Hz), 6.46 (br. s, 1H, H$_h$, H$_{h'}$ or H$_{h''}$), 6.19 (br. s, 2H, H$_k$, H$_{k'}$ or H$_{k''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 5.64 (br. s, 1H, H$_l$, H$_{l'}$ or H$_{l''}$) **^{13}C {^{1}H} NMR** (d_6-DMSO, 100.6 MHz, 300.0 K) δ 169.5 (quaternary C-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.0 (quaternary C-O, C$_f$, C$_{f'}$ or C$_{f''}$), 168.5 (quaternary C-O, C$_f$, C$_{f'}$ or C$_{f''}$), 156.5 (quaternary aniline C, C$_e$, C$_{e'}$ or C$_{e''}$ and C$_e$, C$_{e'}$ or C$_{e''}$), 155.3 (quaternary aniline C, C$_e$, C$_{e'}$ or C$_{e''}$), 151.5 (quaternary aromatic C, C$_i$, C$_{i'}$ and C$_{i''}$), 150.0 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$ and C$_a$, C$_{a'}$ or C$_{a''}$), 149.5 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$), 146.3 (quaternary aromatic C, C$_g$, C$_{g'}$ and C$_{g''}$), 141.4 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 140.5 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 140.4 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 129.7 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$), 129.4 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$), 128.4 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$), 128.2 (aniline CH, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 128.1 (aniline CH, C$_d$, C$_{d'}$ or C$_{d''}$), 126.1 (aromatic CH, C$_l$, C$_{l'}$ or C$_{l''}$ and C$_l$, C$_{l'}$ or C$_{l''}$), 125.3 (aromatic CH, C$_l$, C$_{l'}$ or C$_{l''}$), 124.3 (aromatic CH, C$_k$, C$_{k'}$ or C$_{k''}$ and C$_{k'}$ or C$_{k''}$), 123.4 (aromatic CH, C$_k$, C$_{k'}$ or C$_{k''}$), 114.7 (aromatic CH, C$_j$, C$_{j'}$ or C$_{j''}$), 114.4 (aromatic CH, C$_j$, C$_{j'}$ or C$_{j''}$ and C$_j$, C$_{j'}$ or C$_{j''}$), 111.2 (aromatic CH, C$_h$, C$_{h'}$ or C$_{h''}$), 111.1 (aromatic CH, C$_h$, C$_{h'}$ or C$_{h''}$), 111.0 (aromatic CH, C$_h$, C$_{h'}$ or C$_{h''}$) **Analysis calculated** C 61.37, H 3.43, N 11.93 % **Analysis found** C 61.19, H 3.51, N 11.73 % **ES MS (+)** (CH$_3$OH) *m/z* 705.13 [MH$^+$] **IR** (cm^{-1}) ν 3079 (w, ArCH), 1625 (m, CO), 1592 (s, CO), 1483 (m), 1366 (m), 1262 (w), 1187 (w), 1118 (w), 1005 (w), 764 (m), 680 (m)

8.7.4 Synthesis of CoC$_{36}$H$_{24}$N$_6$O$_3$F$_3$ (2.4)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a red solid (0.255 g, 0.362 mmol, 86 %). Recrystallisation by vapour diffusion (methanol/diethyl ether) at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
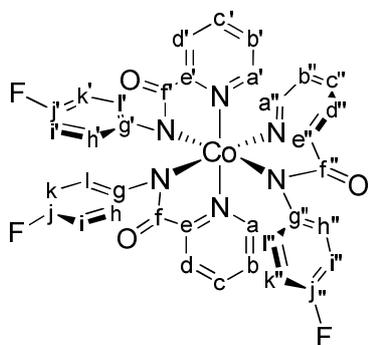
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.1 K) δ 9.09 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.52 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.36 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.16 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 8.00 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.87 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz and ^3J (^{1}H-^{1}H) = 1.2 Hz), 7.80 (m, 2H, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.69 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 6.8 Hz, ^3J (^{1}H-^{1}H) = 5.5 Hz and ^4J (^{1}H-^{1}H) = 1.5 Hz), 7.23 (ddd, 2H, H$_b$, H$_{b'}$ or H$_{b''}$ and H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 6.8 Hz, ^3J (^{1}H-^{1}H) = 5.2 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.15 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 6.97 (t, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 8.0 Hz), 6.90 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.66 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.46 (t, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 8.6 Hz), 6.09 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.0 K) δ 170.6 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.8 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.1 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 158.1 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.8 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.4 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 149.4 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.8 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.3 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 141.7 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 140.7 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$ and C$_j$, C$_{j'}$ or C$_{j''}$), 139.9 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.7 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.5 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 128.9 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 128.1 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 127.1 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.8 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.6 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.7 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 116.4 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$, and C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 116.2 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 115.8 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ and C$_{g''}$), 115.1 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ and C$_{i''}$ and C$_{k''}$), 114.9 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ and C$_{i''}$ and C$_{k''}$, and C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ and C$_{i''}$ and C$_{k''}$) **Analysis calculated** (+ 1 ½ CH$_2$Cl$_2$) C 54.14, H 3.27, N 10.10 % **Analysis found** C 54.60, H 3.40, N 10.20 % **ES MS (+)** (CH$_3$OH) *m/z* 705.13 [MH$^+$] **IR** (cm^{-1}) v 3064 (w, ArCH), 1620 (m, CO), 1585 (s, CO), 1567 (m), 1499 (s), 1367 (m), 1211 (m), 1151 (m), 1094 (w), 826 (m), 788 (m), 682 (m), 529 (m)

8.7.5 Synthesis of CoC$_{36}$H$_{21}$N$_6$O$_3$F$_6$ (2.5)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a brown solid (0.245 g, 0.323 mmol, 77 %). Recrystallisation by vapour diffusion (chloroform/diethyl ether) at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
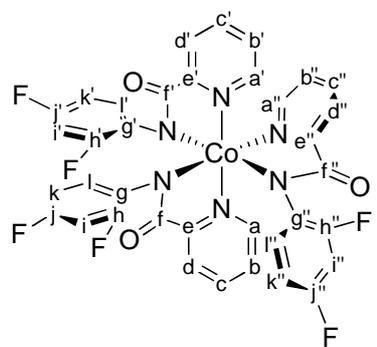
^{1}H NMR (CDCl$_3$, 500.23 MHz, 299.9 K) δ 9.17 (m, 1H, H$_a$, H$_{a'}$ or H$_{a''}$), 8.74 (t, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 6.0 Hz), 8.32 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 8.10 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz and ^3J (^{1}H-^{1}H) = 1.2 Hz), 8.04 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.94 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz and ^3J (^{1}H-^{1}H) = 1.0 Hz), 7.83 (m, 2H, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.59 (m, 2H, H$_a$, H$_{a'}$ or H$_{a''}$ and H$_b$, H$_{b'}$ or H$_{b''}$), 7.47 (m, 1H, H$_k$, H$_{k'}$ or H$_{k''}$), 7.29 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.21 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 6.98 (ddd, 1H, H$_i$, H$_{i'}$ or H$_{i''}$, ^3J (^{1}H-^{1}H) = 11.1 Hz, ^3J (^{1}H-^{1}H) = 8.4 Hz and ^4J (^{1}H-^{1}H) = 2.7 Hz), 6.70 (td, 1H, H$_k$, H$_{k'}$ or H$_{k''}$, ^3J (^{1}H-^{1}H) = 8.2 Hz and ^3J (^{1}H-^{1}H) = 2.1 Hz), 6.62 (td, 1H, H$_k$, H$_{k'}$ or H$_{k''}$, ^3J (^{1}H-^{1}H) = 8.2 Hz and ^3J (^{1}H-^{1}H) = 2.4 Hz), 6.31 (m, 3H, H$_i$, H$_{i'}$ or H$_{i''}$, H$_i$, H$_{i'}$ or H$_{i''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 5.37 (m, 2H, H$_l$, H$_{l'}$ or H$_{l''}$ and H$_l$, H$_{l'}$ or H$_{l''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.1 K) δ 169.6 (quaternary C-O, C$_f$, C$_{f'}$ and C$_{f''}$), 157.6 (quaternary aniline C, C$_e$, C$_{e'}$ or C$_{e''}$), 156.1 (quaternary aniline C, C$_e$, C$_{e'}$ or C$_{e''}$), 155.2 (quaternary aniline C, C$_e$, C$_{e'}$ or C$_{e''}$), 150.7 (quaternary aromatic C, C$_h$, C$_{h'}$ or C$_{h''}$), 150.6 (quaternary aromatic C, C$_h$, C$_{h'}$ or C$_{h''}$ and C$_h$, C$_{h'}$ or C$_{h''}$), 149.9 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$), 149.8 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$), 149.7 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$), 140.6 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 140.2 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 139.7 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 133.6 (quaternary aromatic C, C$_j$, C$_{j'}$ or C$_{j''}$), 131.5 (quaternary aromatic C, C$_j$, C$_{j'}$ or C$_{j''}$), 130.0 (quaternary aromatic C, C$_j$, C$_{j'}$ or C$_{j''}$), 128.4 (quaternary aromatic C, C$_g$, C$_{g'}$ or C$_{g''}$), 128.3 (quaternary aromatic C, C$_g$, C$_{g'}$ or C$_{g''}$), 128.2 (quaternary aromatic C, C$_g$, C$_{g'}$ or C$_{g''}$), 127.5 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$), 127.3 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$), 126.7 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$), 126.2 (aniline CH, C$_d$, C$_{d'}$ or C$_{d''}$), 125.5 (aniline CH, C$_d$, C$_{d'}$ or C$_{d''}$), 125.2 (aniline CH, C$_d$, C$_{d'}$ or C$_{d''}$), 112.4 (aromatic CH, C$_l$, C$_{l'}$ or C$_{l''}$), 112.2 (aromatic CH, C$_l$, C$_{l'}$ or C$_{l''}$ and C$_l$, C$_{l'}$ or C$_{l''}$), 111.0 (aromatic CH, C$_k$, C$_{k'}$ or C$_{k''}$ and C$_k$, C$_{k'}$ or C$_{k''}$), 110.9 (aromatic CH, C$_k$, C$_{k'}$ or C$_{k''}$), 103.9 (aromatic CH, C$_i$, C$_{i'}$ or C$_{i''}$), 103.7 (aromatic CH, C$_i$, C$_{i'}$ or C$_{i''}$ and C$_i$, C$_{i'}$ or C$_{i''}$) **Analysis calculated** (+ ½ CH$_2$Cl$_2$) C 54.73, H 2.77, N 10.49 % **Analysis found** C 54.93, H 2.48, N 10.60 % **ES MS (+)** (CH$_3$OH) *m/z* 759.10 [MH$^+$] **IR** (cm^{-1}) ν 3048 (w, ArCH), 1626 (m, CO), 1592 (s, CO), 1497 (s), 1355 (m), 1260 (m), 1143 (m), 966 (m), 847 (m), 807 (m), 765 (m), 682 (m), 541 (m), 511 (m)

8.7.6 Synthesis of $CoC_{36}H_{21}N_6O_3F_6$ (2.6)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a red solid (0.269 g, 0.355 mmol, 84 %). Recrystallisation by slow evaporation (ethanol/water) at room temperature yielded orange crystals suitable for single crystal X-ray diffraction.
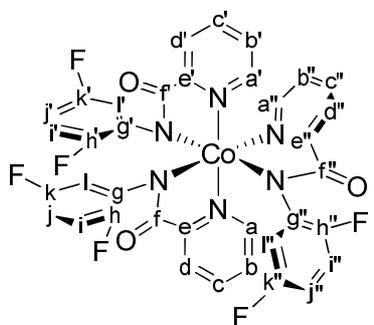
^{1}H NMR (CDCl$_3$, 400.13 MHz, 300.0 K) δ 9.17 (m, 1H, H$_a$, H$_{a'}$ or H$_{a''}$), 8.86 (t, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 6.0 Hz), 8.33 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 8.11 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz and ^3J (^{1}H-^{1}H) = 1.3 Hz), 8.01 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz), 7.94 (m, 2H, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.83 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.2 Hz), 7.60 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.5 Hz and ^4J (^{1}H-^{1}H) = 1.1 Hz), 7.52 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 6.7 Hz, ^3J (^{1}H-^{1}H) = 5.3 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.28 (m, 4H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_l$, H$_{l'}$ or H$_{l''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 6.98 (m, 1H, H$_i$, H$_{i'}$ or H$_{i''}$), 6.69 (m, 1H, H$_i$, H$_{i'}$ or H$_{i''}$), 6.52 (m, 3H, H$_i$, H$_{i'}$ or H$_{i''}$, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 5.26 (ddd, 1H, H$_j$, H$_{j'}$ or H$_{j''}$, ^3J (^{1}H-^{1}H) = 9.6 Hz, ^3J (^{1}H-^{1}H) = 6.4 Hz and ^4J (^{1}H-^{1}H) = 2.9 Hz), 4.99 (ddd, 1H, H$_j$, H$_{j'}$ or H$_{j''}$, ^3J (^{1}H-^{1}H) = 9.1 Hz, ^3J (^{1}H-^{1}H) = 6.1 Hz and ^4J (^{1}H-^{1}H) = 3.0 Hz) **^{13}C {^{1}H} NMR** (d$_6$-DMSO, 100.6 MHz, 300.0 K) δ 169.6 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.0 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 168.4 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 158.8 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 157.1 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 156.0 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 154.8 (quaternary aromatic C̲, C$_k$, C$_{k'}$ or C$_{k''}$), 153.7 (quaternary aromatic C̲, C$_k$, C$_{k'}$ or C$_{k''}$ and C$_k$, C$_{k'}$ or C$_{k''}$), 149.9 (aniline C̲H, C$_a$, C$_{a'}$ and C$_{a''}$), 149.6 (quaternary aromatic C̲, C$_h$, C$_{h'}$ or C$_{h''}$), 141.7 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 141.3 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 141.0 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 128.6 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 128.4 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 128.1 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.1 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.3 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 124.6 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 116.5 (aromatic C̲H, C$_i$, C$_{i'}$ or C$_{i''}$), 115.6 (aromatic C̲H, C$_i$, C$_{i'}$ or C$_{i''}$ and C$_i$, C$_{i'}$ or C$_{i''}$), 114.6 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$ and C$_g$, C$_{g'}$ or C$_{g''}$), 114.3 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 113.1 (quaternary aromatic C̲, C$_j$, C$_{j'}$ or C$_{j''}$), 112.8 (quaternary aromatic C̲, C$_j$, C$_{j'}$ or C$_{j''}$ and C$_j$, C$_{j'}$ or C$_{j''}$), 111.3 (aromatic C̲H, C$_l$, C$_{l'}$ and C$_{l''}$) **Analysis calculated** (+ ½ CH$_2$Cl$_2$) C 54.73, H 2.77, N 10.49 % **Analysis found** C 54.63, H 3.17, N 10.16 % **ES MS (+)** (CH$_3$OH) m/z 759.10 [MH$^+$] **IR** (cm^{-1}) ν 3067 (w, ArCH), 1633 (s, CO), 1597 (s, CO), 1490 (s), 1357 (s), 1242 (m), 1178 (m), 973 (m), 859 (m), 749 9 (s), 684 (m), 592 (w)

8.7.7 Synthesis of CoC$_{36}$H$_{24}$N$_6$O$_3$Cl$_3$ (2.7)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a brown solid (0.129 g, 0.170 mmol, 41 %). Recrystallisation by vapour diffusion (methanol/diethyl ether) at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
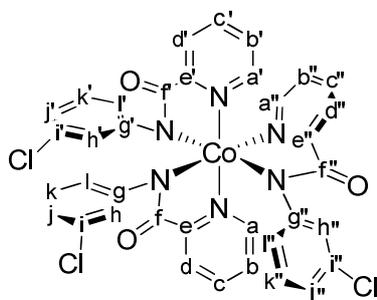
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.1 K) δ 9.04 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.57 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.38 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.17 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.4 Hz), 8.02 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.89 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.4 Hz), 7.82 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.4 Hz), 7.77 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.71 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 6.8 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.36 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 6.8 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.31 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 6.8 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.26 (d, 1H, H$_j$, H$_{j'}$ or H$_{j''}$, ^3J (^{1}H-^{1}H) = 8.0 Hz), 7.23 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 7.17 (m, 2H, H$_h$, H$_{h'}$ or H$_{h''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 7.02 (t, 1H, H$_k$, H$_{k'}$ or H$_{k''}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 6.95 (d, 1H, H$_j$, H$_{j'}$ or H$_{j''}$, ^3J (^{1}H-^{1}H) = 8.0 Hz), 6.84 (d, 1H, H$_l$, H$_{l'}$ or H$_{l''}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 6.75 (t, 1H, H$_k$, H$_{k'}$ or H$_{k''}$, ^3J (^{1}H-^{1}H) = 8.0 Hz), 6.58 (br. s, 2H, H$_h$, H$_{h'}$ or H$_{h''}$ and H$_j$, H$_{j'}$ or H$_{j''}$), 6.37 (br. s, 1H, H$_l$, H$_{l'}$ or H$_{l''}$), 6.18 (br. s, 1H, H$_h$, H$_{h'}$ or H$_{h''}$), 5.89 (br. s, 1H, H$_l$, H$_{l'}$ or H$_{l''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.0 K) δ 170.3 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.6 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 168.7 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 157.6 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 156.5 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 156.2 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 149.3 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.9 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.4 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 147.1 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 145.3 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 145.0 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 140.9 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 140.1 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.9 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 134.9 (quaternary aromatic C̲, C$_i$, C$_{i'}$ or C$_{i''}$), 133.8 (quaternary aromatic C̲, C$_i$, C$_{i'}$ or C$_{i''}$), 133.6 (quaternary aromatic C̲, C$_i$, C$_{i'}$ or C$_{i''}$), 130.4 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$ and C$_j$, C$_{j'}$ or C$_{j''}$), 130.2 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$), 129.0 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 128.3 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 127.8 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 127.2 (aromatic C̲H, C$_k$, C$_{k'}$ or C$_{k''}$ and C$_k$, C$_{k'}$ or C$_{k''}$), 127.1 (aromatic C̲H, C$_k$, C$_{k'}$ or C$_{k''}$), 126.9 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.3 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 125.8 (aromatic C̲H, C$_h$, C$_{h'}$ and C$_{h''}$), 125.3 (aromatic C̲H, C$_l$, C$_{l'}$ or C$_{l''}$), 125.1 (aromatic C̲H, C$_l$, C$_{l'}$ or C$_{l''}$ and C$_l$, C$_{l'}$ or C$_{l''}$) **Analysis calculated** (+ ½ CHCl$_3$) C 53.88, H 3.04, N 10.33 % **Analysis found** C 53.95, H 3.09, N 10.36 % **ES MS (+)** (CH$_3$OH) *m/z* 755.04 [MH$^+$] **IR**

(cm^{-1}) v 3067 (w, ArCH), 1624 (s, CO), 1598 (s, CO), 1471 (m), 1362 (m), 1151 (w), 999 (w), 881 (w), 763 (m), 685 (m), 506 (w)

8.7.8 Synthesis of CoC$_{36}$H$_{24}$N$_6$O$_3$Cl$_3$ (2.8)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a red solid (0.239 g, 0.317 mmol, 75 %). Recrystallisation by vapour diffusion (chloroform/pentane) at 5 °C yielded orange crystals suitable for single crystal X-ray diffraction.
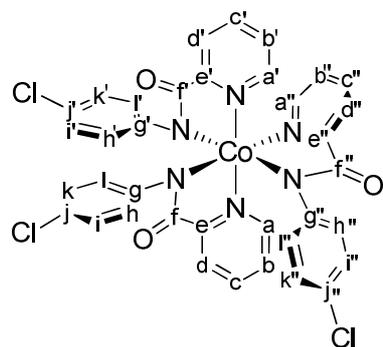
^{1}H NMR (CDCl$_3$, 500.23 MHz, 299.9 K) δ 9.08 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 4.8 Hz), 8.53 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 4.8 Hz), 8.39 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 8.18 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 8.03 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 7.90 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 7.82 (m, 2H, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.70 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.3 Hz), 7.25 (m, 4H, H$_b$, H$_{b'}$ or H$_{b''}$, H$_b$, H$_{b'}$ or H$_{b''}$, and H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$), 7.17 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 4.8 Hz), 6.93 (m, 4H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$or H$_{h''}$ and H$_{l''}$, and H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$), 6.74 (d, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, ^3J (^{1}H-^{1}H) = 8.2 Hz), 6.42 (br. s, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$), 6.07 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.1 K) δ 170.4 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.6 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 168.9 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 157.9 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 157.3 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.7 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 149.4 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.9 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.3 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 142.6 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 142.2 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$ and C$_j$, C$_{j'}$ or C$_{j''}$), 140.8 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 140.0 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.8 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 131.5 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ and C$_{g''}$), 129.6 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 129.0 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$, and C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 128.3 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ and C$_{i''}$ and C$_{k''}$), 127.3 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$ and C$_b$, C$_{b'}$ or C$_{b''}$), 126.9 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.3 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 125.9 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$) **Analysis calculated** (+ CH$_2$Cl$_2$) C 52.98, H 3.12, N 10.02 % **Analysis found** C 52.70, H 2.75, N 9.50 % **ES MS (+)** (CH$_3$OH) *m/z* 755.04 [MH$^+$] **IR** (cm^{-1}) v 3085 (w, ArCH), 1617 (m, CO), 1593 (s, CO), 1566 (s, CO), 1482 (s), 1370 (m), 1084 (m), 1008 (m), 816 (m), 764 (m), 705 (m), 508 (m)

8.7.9 Synthesis of CoC$_{36}$H$_{24}$N$_6$O$_3$Br$_3$ (2.9)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a brown solid (0.157 g, 0.177 mmol, 42 %). Recrystallisation by vapour diffusion (methanol/diethyl ether) at 5 °C yielded red crystals suitable for single crystal X-ray diffraction.
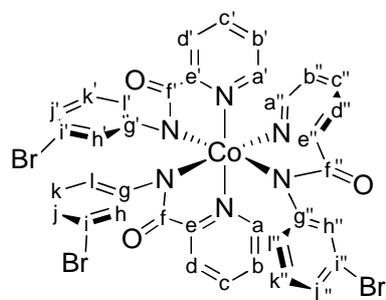
^{1}H NMR (CDCl$_3$, 500.23 MHz, 299.9 K) δ 9.03 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.57 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.39 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.17 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz and ^3J (^{1}H-^{1}H) = 1.0 Hz), 8.01 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.90 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 0.9 Hz), 7.83 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz and ^3J (^{1}H-^{1}H) = 1.0 Hz), 7.77 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.71 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.4 Hz), 7.39 (m, 2H, H$_b$, H$_{b'}$ or H$_{b''}$ and H$_j$, H$_{j'}$ or H$_{j''}$), 7.34 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.4 Hz), 7.24 (m, 2H, H$_a$, H$_{a'}$ or H$_{a''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 7.10 (m, 2H, H$_h$, H$_{h'}$ or H$_{h''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 6.98 (m, 2H, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 6.72 (m, 3H, H$_h$, H$_{h'}$ or H$_{h''}$, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 6.47 (br. s, 1H, H$_l$, H$_{l'}$ or H$_{l''}$), 6.22 (br. s, 1H, H$_h$, H$_{h'}$ or H$_{h''}$), 6.06 (br. s, 1H, H$_l$, H$_{l'}$ or H$_{l''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.0 K) δ 170.3 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f'}$), 169.1 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f'}$), 168.7 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f'}$), 157.6 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e'}$), 156.6 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e'}$), 156.2 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e'}$), 149.3 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.9 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.4 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 147.4 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 145.5 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 145.3 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 140.9 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 140.1 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 140.0 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 131.2 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$), 130.7 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$), 130.5 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$), 129.4 (aromatic C̲H, C$_h$, C$_{h'}$ or C$_{h''}$), 129.2 (aromatic C̲H, C$_h$, C$_{h'}$ or C$_{h''}$ and C$_h$, C$_{h'}$ or C$_{h''}$), 128.4 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 128.2 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 128.0 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 127.3 (aromatic C̲H, C$_k$, C$_{k'}$ or C$_{k''}$ and C$_k$, C$_{k'}$ or C$_{k''}$), 127.2 (aromatic C̲H, C$_k$, C$_{k'}$ or C$_{k''}$), 126.9 (aromatic C̲H, C$_l$, C$_{l'}$ and C$_{l''}$), 126.4 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.0 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.8 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 123.0 (quaternary aromatic C̲, C$_i$, C$_{i'}$ or C$_{i''}$ and C$_i$, C$_{i'}$ or C$_{i''}$), 121.7 (quaternary aromatic C̲, C$_i$, C$_{i'}$ or C$_{i''}$) **Analysis calculated** C 48.73, H 2.73, N 9.47 % **Analysis found** C 48.64, H 2.69, N 9.57 % **ES MS (+)** (CH$_3$OH) *m/z* 888.88 [MH$^+$] **IR** (cm^{-1}) ν 3063 (w, ArCH), 1624 (s, CO), 1593 (s, CO), 1468 (m), 1363 (m), 1293 (w), 1148 (w), 1069 (w), 953 (w), 861 (m), 763 (s), 722 (m), 682 (s), 560 (m)

8.7.10 Synthesis of CoC$_{36}$H$_{24}$N$_6$O$_3$Br$_3$ (2.10)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a red solid (0.250 g, 0.282 mmol, 67 %). Recrystallisation by vapour diffusion (methanol/diethyl ether) at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
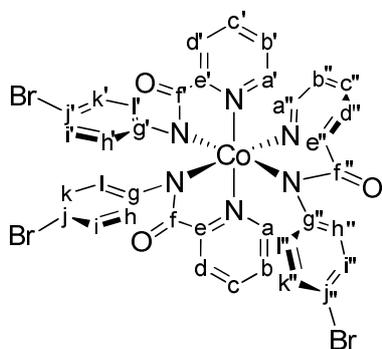
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.2 K) δ 9.07 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 4.7 Hz), 8.52 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 4.8 Hz), 8.39 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.18 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 8.03 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 7.91 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.4 Hz), 7.83 (m, 2H, H$_b$, H$_{b'}$ or H$_{b''}$ and H$_c$, H$_{c'}$ or H$_{c''}$), 7.71 (m, 2H, H$_b$, H$_{b'}$ or H$_{b''}$ and H$_b$, H$_{b'}$ or H$_{b''}$), 7.51 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.9 Hz), 7.39 (d, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 8.0 Hz), 7.17 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 4.6 Hz), 7.08 (m, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.89 (d, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, ^3J (^{1}H-^{1}H) = 8.0 Hz), 6.82 (br. s, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$), 6.36 (br. s, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$), 6.00 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$) **^{13}C {^{1}H}** **NMR** (CDCl$_3$, 125.8 MHz, 299.9 K) δ 170.4 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.7 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.5 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 157.9 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 157.1 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.3 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 149.4 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.9 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.3 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 140.8 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 140.0 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.9 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 132.5 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ and C$_{j''}$), 132.0 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 131.9 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 131.3 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 129.3 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ and C$_{i''}$ and C$_{k''}$, and C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ and C$_{i''}$ and C$_{k''}$), 128.32 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ and C$_{i''}$ and C$_{k''}$), 127.3 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$ and C$_b$, C$_{b'}$ or C$_{b''}$), 126.9 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.4 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 125.9 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 121.2 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 119.4 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ and C$_{g''}$, and C$_g$, C$_{g'}$ and C$_{g''}$) **Analysis calculated** (+ 1 CH$_2$Cl$_2$) C 45.71, H 2.70, N 8.64 % **Analysis found** C 45.55, H 2.72, N 8.83 % **ES MS (+)** (CH$_3$OH) *m/z* 888.88 [MH$^+$] **IR** (cm^{-1}) v 3069 (w, ArCH), 1615 (m, CO), 1592 (s, CO), 1566 (m), 1478 (m), 1362 (m), 1260 (m), 1070 (w), 1004 (w), 811 (m), 763 (m), 685 (m), 507 (m)

8.7.11 Synthesis of CoC$_{39}$H$_{24}$N$_6$O$_3$F$_9$ (2.11)

Purified by column chromatography (methanol/diethyl ether 4:1) to give a red solid (0.190 g, 0.222 mmol, 53 %). Recrystallisation by vapour diffusion (chloroform/pentane) at room temperature yielded orange crystals suitable for single crystal X-ray diffraction.
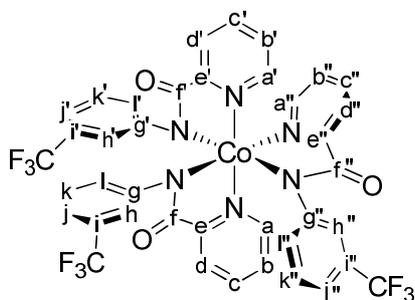
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.0 K) δ 9.09 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.67 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.45 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.23 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.94 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 7.89 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.4 Hz), 7.78 (m, 3H, H$_b$, H$_{b'}$ or H$_{b''}$, H$_c$, H$_{c'}$ or

H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.63 (d, 1H, H$_j$, H$_{j'}$ or H$_{j''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 7.52 (d, 1H, H$_j$, H$_{j'}$ or H$_{j''}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 7.31 (m, 7H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_h$, H$_{h'}$ or H$_{h''}$, H$_k$, H$_{k'}$ or H$_{k''}$, H$_k$, H$_{k'}$ or H$_{k''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 7.12 (d, 1H, H$_l$, H$_{l'}$ or H$_{l''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.03 (t, 1H, H$_k$, H$_{k'}$ or H$_{k''}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 6.86 (m, 2H, H$_h$, H$_{h'}$ or H$_{h''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 6.68 (br. s, 1H, H$_h$, H$_{h'}$ or H$_{h''}$), 6.21 (br. s, 1H, H$_l$, H$_{l'}$ or H$_{l''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.1 K) δ 170.3 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.7 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 168.8 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 157.7 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.6 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.2 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 149.0 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.3 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 146.4 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 144.6 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$ and C$_g$, C$_{g'}$ or C$_{g''}$), 144.3 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 141.1 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 140.1 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 140.0 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 132.9 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$), 132.1 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$), 131.6 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$), 130.8 ($\underline{C}$F$_3$), 130.0 ($\underline{C}$F$_3$ and $\underline{C}$F$_3$), 128.8 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 128.3 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 128.0 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 127.6 (aromatic $\underline{C}$H, C$_k$, C$_{k'}$ or C$_{k''}$ and C$_k$, C$_{k'}$ or C$_{k''}$), 127.1 (aromatic $\underline{C}$H, C$_k$, C$_{k'}$ or C$_{k''}$), 126.4 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 126.0 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 124.6 (aromatic $\underline{C}$H, C$_l$, C$_{l'}$ or C$_{l''}$), 123.6 (aromatic $\underline{C}$H, C$_l$, C$_{l'}$ or C$_{l''}$), 122.8 (aromatic $\underline{C}$H, C$_l$, C$_{l'}$ or C$_{l''}$), 122.3 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$ or C$_{h''}$ and C$_h$, C$_{h'}$ or C$_{h''}$), 122.1 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$ or C$_{h''}$), 121.9 (quaternary aromatic $\underline{C}$, C$_i$, C$_{i'}$ or C$_{i''}$ and C$_i$, C$_{i'}$ or C$_{i''}$), 121.6 (quaternary aromatic $\underline{C}$, C$_i$, C$_{i'}$ or C$_{i''}$) **Analysis calculated** (+ ½ CH$_2$Cl$_2$) C 52.89, H 2.81, N 9.37 % **Analysis found** C 52.96, H 2.73, N 9.33 % **ES MS (+)** (CH$_3$OH) m/z 855.12 [MH$^+$] **IR** (cm^{-1}) ν 3081 (w, ArCH), 1629 (m, CO), 1596 (m, CO), 1434 (m), 1362 (m), 1323 (s), 1117 (s), 1092 (m), 1066 (m), 797 (m), 776 (m), 684 (m), 657 (m)

8.7.12 Synthesis of CoC$_{39}$H$_{24}$N$_6$O$_3$F$_9$ (2.12)

Purified by column chromatography (methanol/diethyl ether 4:1) to give a red solid (0.183 g, 0.214 mmol, 51 %). Recrystallisation by vapour diffusion (DCM/diethyl ether) at room temperature yielded orange crystals suitable for single crystal X-ray diffraction.
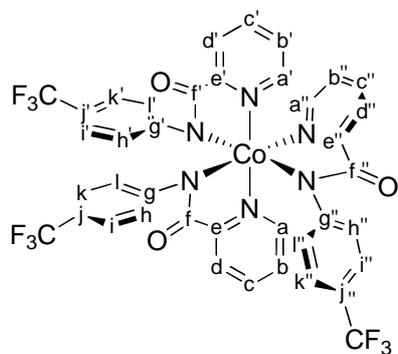
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.1 K) δ 9.10 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.56 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.4 Hz), 8.44 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.1 Hz), 8.23 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz), 8.00 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 6.9 Hz), 7.88 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.1 Hz), 7.83 (1H, t, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.1 Hz), 7.76 (m, 2H, H$_a$, H$_{a'}$ or H$_{a''}$ and H$_b$, H$_{b'}$ or H$_{b''}$), 7.56 (d, 2H, H$_i$, H$_{i'}$ or H$_{i''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 7.29 (m, 5H, H$_b$, H$_{b'}$ or H$_{b''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_d$, H$_{d'}$ or H$_{d''}$, H$_i$, H$_{i'}$ or H$_{i''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 7.09 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 7.00 (d, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 8.0 Hz), 6.66 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.20 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$) **^{13}C {^{1}H} NMR** (d_6-DMSO, 100.6 MHz, 300.0 K) δ 169.3 (quaternary C-O, C$_f$, C$_{f'}$ or C$_{f'}$), 169.2 (quaternary C-O, C$_f$, C$_{f'}$ or C$_{f'}$), 168.5 (quaternary C-O, C$_f$, C$_{f'}$ or C$_{f'}$), 166.2 (quaternary aniline C, C$_e$, C$_{e'}$ or C$_{e''}$), 161.0 (quaternary aniline C, C$_e$, C$_{e'}$ or C$_{e''}$), 156.0 (quaternary aniline C, C$_e$, C$_{e'}$ or C$_{e''}$), 155.1 (quaternary aromatic C, C$_g$, C$_{g'}$ or C$_{g''}$), 154.9 (quaternary aromatic C, C$_g$, C$_{g'}$ or C$_{g''}$), 151.4 (quaternary aromatic C, C$_g$, C$_{g'}$ or C$_{g''}$), 150.1 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$), 150.0 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$), 149.8 (aniline CH, C$_a$, C$_{a'}$ or C$_{a''}$), 148.9 (quaternary aromatic C, C$_j$, C$_{j'}$ and C$_{j''}$), 148.5 (quaternary aromatic C, C$_j$, C$_{j'}$ and C$_{j''}$ and C$_j$, C$_{j'}$ and C$_{j''}$), 141.7 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 140.7 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 140.6 (aniline CH, C$_c$, C$_{c'}$ or C$_{c''}$), 129.6 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$), 129.0 (aniline CH, C$_b$, C$_{b'}$ or C$_{b''}$ and C$_b$, C$_{b'}$ or C$_{b''}$), 128.7 (CF$_3$, CF$_3$ and CF$_3$), 128.4 (aromatic CH, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 128.0 (aromatic CH, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$, and C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 126.4 (aniline CH, C$_d$, C$_{d'}$ or C$_{d''}$), 125.6 (aniline CH, C$_d$, C$_{d'}$ or C$_{d''}$), 125.5 (aniline CH, C$_d$, C$_{d'}$ or C$_{d''}$), 124.9 (aromatic CH, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 124.3 (aromatic CH, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 124.2 (aromatic CH, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$) **Analysis calculated** (+ ½ H$_2$O + 2 ½ CH$_2$Cl$_2$) C 46.68, H 2.08, N 7.87 % **Analysis found** C 46.50, H 2.00, N 7.80 % **ES MS (+)** (CH$_3$OH) *m/z* 855.12 [MH$^+$] **IR** (cm^{-1}) ν 3042 (w, ArCH), 1631 (m, CO), 1598 (m, CO), 1362 (m), 1321 (s), 1107 (m), 1066 (m), 825 (w), 760(w), 686 (w), 506 (w)

8.7.13 Synthesis of CoC₄₂H₂₁N₆O₃F₁₈ (2.13)

8.7.13 Synthesis of $CoC_{42}H_{21}N_6O_3F_{18}$ (2.13)

Purified by column chromatography (methanol/diethyl ether 4:1) to give a red solid (0.272 g, 0.257 mmol, 61 %). Recrystallisation by vapour diffusion (chloroform/pentane) at 5 °C yielded orange crystals suitable for single crystal X-ray diffraction.
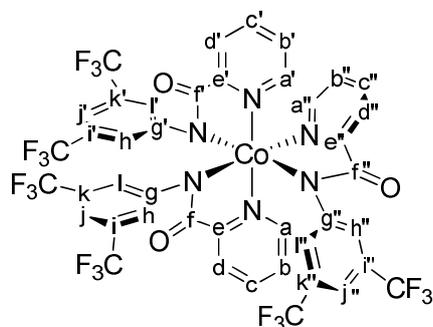
¹H NMR (CDCl₃, 400.13 MHz, 300.0 K) δ 8.98 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.80 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.7 Hz), 8.51 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.2 Hz), 8.31 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.2 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 8.00 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.87 (m, 3H, H$_c$, H$_{c'}$ or H$_{c''}$, and H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 7.80 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.70 (s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 7.50 (m, 5H, H$_b$, H$_{b'}$ or H$_{b''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_d$, H$_{d'}$ or H$_{d''}$, H$_d$, H$_{d'}$ or H$_{d''}$ and H$_j$, H$_{j'}$ or H$_{j''}$), 7.37 (m, 2H, H$_a$, H$_{a'}$ or H$_{a''}$ and H$_j$, H$_{j'}$ or H$_{j''}$), 6.94 (s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.87 (br.s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$) **¹³C {¹H} NMR** (d_6-DMSO, 100.6 MHz, 300.0 K) δ 169.8 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$ and C$_f$, C$_{f'}$ or C$_{f''}$), 168.9 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 154.9 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 154.0 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 153.9 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 150.5 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$ and C$_a$, C$_{a'}$ or C$_{a''}$), 150.3 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 149.1 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 146.3 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$ and C$_g$, C$_{g'}$ or C$_{g''}$), 142.2 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 141.4 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 141.2 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 130.4 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 130.3 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 129.9 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 128.7 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 127.9 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.9 ($\underline{C}$F₃, $\underline{C}$F₃ and $\underline{C}$F₃), 125.8 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$ and C$_j$, C$_{j'}$ or C$_{j''}$), 125.4 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$), 124.2 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 123.3 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 121.4 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 118.5 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 117.6 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 117.3 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$) **Analysis calculated** (+ ½ CHCl₃) C 45.65, H 1.94, N 7.52 % **Analysis found** C 45.69, H 1.66, N 7.52 % **ES MS (+)** (CH₃OH) *m/z* 1059.08 [MH⁺] **IR** (cm⁻¹) ν 3087 (w, ArCH), 1635 (m, CO), 1601 (m, CO), 1465 (w), 1369 (m), 1273 (s), 1058 (s), 967 (w), 897 (w), 762 (w), 682 (m)

8.7.14 Synthesis of CoC$_{39}$H$_{33}$N$_6$O$_3$ (2.14a)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a red solid (0.206 g, 0.297 mmol, 71 %). Recrystallisation by vapour diffusion (methanol/diethyl ether) at 5 °C yielded red crystals suitable for single crystal X-ray diffraction.
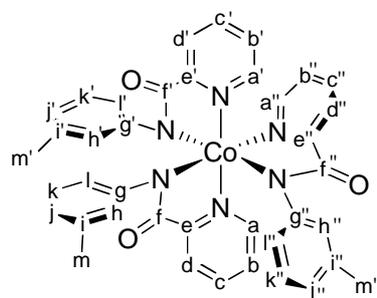
^{1}H NMR (CDCl$_3$, 400.13 MHz, 298.0 K) δ 9.17 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.4 Hz), 8.57 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.4 Hz), 8.34 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.11 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 8.00 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.75 (m, 3H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.66 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) =

6.7 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.12 (m, 6H, H$_b$, H$_{b'}$ or H$_{b''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_c$, H$_{c'}$ or H$_{c''}$, H$_h$, H$_{h'}$ or H$_{h''}$, H$_h$, H$_{h'}$ or H$_{h''}$ and H$_j$, H$_{j'}$ or H$_{j''}$), 6.86 (br. s, 2H, H$_k$, H$_{k'}$ or H$_{k''}$ and H$_j$, H$_{j'}$ or H$_{j''}$), 6.75 (m, 2H, H$_h$, H$_{h'}$ or H$_{h''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 6.62 (m, 2H, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 6.12 (br. s, 2H, H$_k$, H$_{k'}$ or H$_{k''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 5.75 (br. s, 1H, H$_l$, H$_{l'}$ or H$_{l''}$), 2.25 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 2.08 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 1.95 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 170.3 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.4 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 168.7 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 158.7 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 157.2 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.8 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 149.7 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.9 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.4 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 145.8 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 144.1 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 143.6 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 140.4 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.3 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.2 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 138.7 (quaternary aromatic $\underline{C}$, C$_i$, C$_{i'}$ or C$_{i''}$), 137.3 (quaternary aromatic $\underline{C}$, C$_i$, C$_{i'}$ or C$_{i''}$ and C$_i$, C$_{i'}$ or C$_{i''}$), 129.1 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$ or C$_{h''}$), 128.7 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$ or C$_{h''}$), 128.4 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$ or C$_{h''}$), 127.7 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.6 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$ and C$_b$, C$_{b'}$ or C$_{b''}$), 126.6 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.4 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.3 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.1 (aromatic $\underline{C}$H, C$_k$, C$_{k'}$ or C$_{k''}$), 125.9 (aromatic $\underline{C}$H, C$_k$, C$_{k'}$ or C$_{k''}$ and C$_k$, C$_{k'}$ or C$_{k''}$), 125.7 (aromatic $\underline{C}$H, C$_l$, C$_{l'}$ or C$_{l''}$), 125.5 (aromatic $\underline{C}$H, C$_l$, C$_{l'}$ or C$_{l''}$ and C$_l$, C$_{l'}$ or C$_{l''}$), 125.1 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$ and C$_j$, C$_{j'}$ or C$_{j''}$), 124.5 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$), 21.4 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$), 21.0 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$), 20.8 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$) **Analysis calculated** (+ ½ CHCl$_3$) C 63.06, H 4.49, N 11.17 % **Analysis found** C 63.22, H 4.66, N 11.34 % **ES MS (+)** (CH$_3$OH) m/z 693.20 [MH$^+$] **IR** (cm^{-1}) ν 3036 (w, ArCH), 2916 (w, MeCH), 1620 (m, CO), 1590 (s, CO), 1565 (s, CO), 1483 (m), 1365 (m), 1294 (w), 1093 (w), 764 (m), 682 (m)

8.7.15 Synthesis of CoC$_{39}$H$_{33}$N$_6$O$_3$ (2.15a)

Purified by column chromatography (methanol/diethyl ether 3:2) to give a red solid (0.141 g, 0.203 mmol, 48 %). Recrystallisation by vapour diffusion (methanol/diethyl ether) at 5 °C yielded red crystals suitable for single crystal X-ray diffraction.
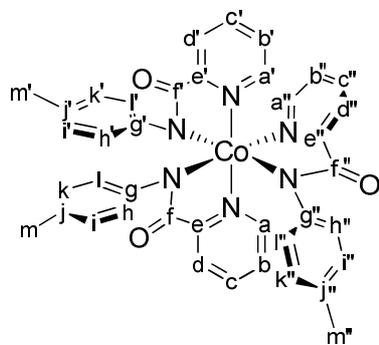
^{1}H NMR (CDCl$_3$, 400.13 MHz, 298.0 K) δ 9.15 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.4 Hz), 8.58 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.4 Hz), 8.35 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.11 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.96 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.75 (m, 3H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_c$, H$_{c'}$ or H$_{c''}$), 7.65 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 6.7 Hz, ^3J (^{1}H-^{1}H) = 5.6 Hz and ^4J (^{1}H-^{1}H) = 1.3 Hz), 7.14 (m, 3H, H$_b$, H$_{b'}$ or H$_{b''}$, H$_b$, H$_{b'}$ or H$_{b''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.06 (d, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 6.84 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.75 (d, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz), 6.52 (d, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 8.1 Hz), 6.33 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.02 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 2.26 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 2.08 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 1.98 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 170.5 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.6 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 167.0 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 158.6 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 157.2 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.8 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 149.7 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 149.0 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.3 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 143.1 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 141.5 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 141.0 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 140.3 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.2 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.1 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 135.3 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 134.6 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 134.3 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 130.0 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 129.4 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 128.7 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 127.6 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 127.2 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$ and C$_b$, C$_{b'}$ or C$_{b''}$), 126.6 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.5 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.4 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.8 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 125.4 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$, and C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 21.1 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$), 21.0 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$), 20.9 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$) **Analysis calculated** (+ ⅔ CH$_2$Cl$_2$) C 63.58, H 4.62, N 11.22 % **Analysis found** C 63.10, H 4.50, N 10.90 % **ES MS (+)** (CH$_3$OH) *m/z* 693.20 [MH$^+$] **IR** (cm^{-1}) ν 3052 (w, ArCH), 2920 (w, MeCH), 1622 (m, CO), 1589 (s, CO), 1565 (s, CO), 1506 (m), 1368 (m), 1294 (w), 1149 (w), 1095 (w), 808 (m), 757 (m), 681 (m), 524 (m)

8.7.16 Synthesis of $CoC_{42}H_{39}N_6O_3$ (2.16a)

Purified by column chromatography (methanol/diethyl ether 4:1) to give a red solid (0.210 g, 0.286 mmol, 68 %). Recrystallisation by vapour diffusion (chloroform/pentane) at room temperature yielded orange crystals suitable for single crystal X-ray diffraction.
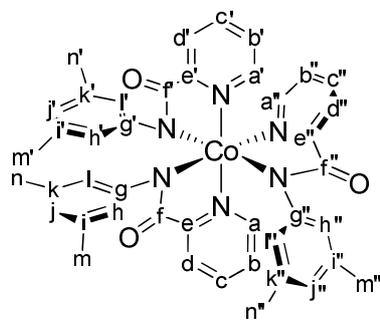
^{1}H NMR (CDCl$_3$, 400.13 MHz, 300.0 K) δ 9.17 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.56 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 8.32 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 8.09 (t, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 8.02 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 7.75 (m, 4H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_c$, H$_{c'}$ or H$_{c''}$, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.65 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.5 Hz and ^4J (^{1}H-^{1}H) = 1.1 Hz), 7.14 (m, 4H, H$_b$, H$_{b'}$ or H$_{b''}$ and H$_b$, H$_{b'}$ or H$_{b''}$, and H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.90 (s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 6.56 (m, 3H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, and H$_j$, H$_{j'}$ or H$_{j''}$), 6.44 (s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 5.77 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 2.19 (br. s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$), 2.05 (s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$), 1.93 (s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 170.2 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.5 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 168.6 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 158.9 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 157.3 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 157.0 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 149.8 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.9 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.4 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 145.7 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 144.1 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 143.6 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 140.3 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.1 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.0 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 137.0 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$, and C$_{i''}$ and C$_{k''}$), 127.6 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 127.5 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.9 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.6 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.4 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.2 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.8 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$, and C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 125.4 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 124.8 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ and C$_{j''}$), 21.4 ($\underline{C}$H$_3$, C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$), 21.0 ($\underline{C}$H$_3$, C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$), 20.8 ($\underline{C}$H$_3$, C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$) **Analysis calculated** (+ ½ CHCl$_3$) C 64.26, H 5.01, N 10.58 % **Analysis found** C 64.30, H 5.50, N 10.80 % **ES MS (+)** (CH$_3$OH) m/z 735.25 [MH$^+$] **IR** (cm^{-1}) ν 3083 (w, ArCH), 2914 (w, MeCH), 1616 (m, CO), 1595 (s, CO), 1581 (s, CO), 1470 (w), 1374 (m), 1278 (w), 1097 (w), 1026 (w), 840 (w), 765 (m), 681 (m)

8.7.17 Synthesis of CoC$_{39}$H$_{33}$N$_6$O$_6$ (2.17a)

Purified by column chromatography (methanol/diethyl ether 4:1) to give a red solid (0.149 g, 0.201 mmol, 48 %). Recrystallisation by slow evaporation from methanol at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
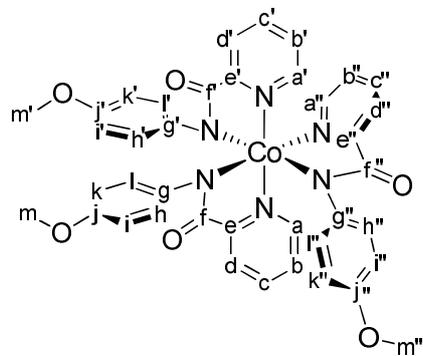
^{1}**H NMR** (CDCl$_3$, 400.13 MHz, 300.0 K) δ 9.12 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.55 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 8.36 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 8.11 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.97 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 7.76 (m, 3H, H$_b$, H$_{b'}$ or H$_{b''}$, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.64 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz, ^3J (^{1}H-^{1}H) = 5.1 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.15 (m, 3H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_b$, H$_{b'}$ or H$_{b''}$ and H$_c$, H$_{c'}$ or H$_{c''}$), 6.88 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.81 (d, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 8.4 Hz), 6.51 (d, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 6.40 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.29 (d, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 8.8 Hz), 6.09 (br. s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 3.76 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 3.66 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 3.58 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$) ^{13}**C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 170.6 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.7 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.2 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 158.5 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 157.4 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 157.2 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.8 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 156.7 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 156.6 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 149.6 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 149.0 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.3 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 140.3 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.4 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.2 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 138.5 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 137.1 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 136.4 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 128.3 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$ and C$_b$, C$_{b'}$ or C$_{b''}$), 127.6 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.6 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 126.5 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$, and C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 125.8 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 125.3 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 114.6 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 114.2 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 113.5 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 55.3 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$ and C$_m$, C$_{m'}$ or C$_{m''}$), 55.2 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$) **Analysis calculated** (+ CH$_2$Cl$_2$) C 58.19, H 4.27, N 10.18 % **Analysis found** C 57.29, H 4.48, N 10.23 % **ES MS (+)** (CH$_3$OH) *m/z* 741.19 [MH$^+$] **IR** (cm^{-1}) ν 3065 (w, ArCH), 2939 (w, MeCH), 2835 (w), 1588 (s, CO), 1503 (s), 1370 (m), 1286 (m), 1240 (s), 1027 (m), 822 (m), 763 (m), 683 (m)

8.7.18 Synthesis of CoC$_{42}$H$_{39}$N$_6$O$_9$ (2.18a)

Purified by column chromatography (methanol/diethyl ether 2:3) to give a red solid
(0.135 g, 0.162 mmol, 39 %).
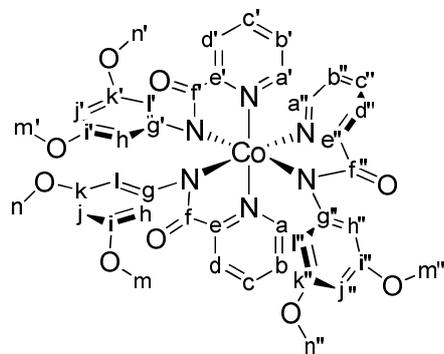
¹H NMR (CDCl$_3$, 400.13 MHz, 300.0 K) δ 9.10 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (¹H-¹H) = 5.1 Hz), 8.63 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (¹H-¹H) = 5.1 Hz), 8.38 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (¹H-¹H) = 7.5 Hz), 8.13 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (¹H-¹H) = 7.4 Hz and ^3J (¹H-¹H) = 1.2 Hz), 7.93 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (¹H-¹H) = 7.5 Hz), 7.82 (m, 2H, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ and H$_{d''}$), 7.75 (td, 1H, H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (¹H-¹H) = 7.4 Hz and ^3J (¹H-¹H) = 1.2 Hz), 7.67 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (¹H-¹H) = 7.0 Hz, ^3J (¹H-¹H) = 5.5 Hz and ^4J (¹H-¹H) = 1.3 Hz), 7.14 (m, 4H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_b$, H$_{b'}$ or H$_{b''}$ and, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.35 (s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 6.17 (br. s, 3H, H$_b$, H$_{b'}$ or H$_{b''}$, and H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.08 (s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 5.98 (s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 5.61 (br.s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 3.63 (s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$), 3.58 (s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$), 3.51 (s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$) **¹³C {¹H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 170.2 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.2 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.1 (quaternary C̲-O, C$_f$, C$_{f'}$ or C$_{f''}$), 161.0 (quaternary aromatic C̲, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 160.6 (quaternary aromatic C̲, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 160.2 (quaternary aromatic C̲, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 158.8 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 157.1 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 156.9 (quaternary aniline C̲, C$_e$, C$_{e'}$ or C$_{e''}$), 150.0 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 149.1 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.9 (aniline C̲H, C$_a$, C$_{a'}$ or C$_{a''}$), 147.8 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 146.3 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 145.6 (quaternary aromatic C̲, C$_g$, C$_{g'}$ or C$_{g''}$), 140.5 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.4 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.2 (aniline C̲H, C$_c$, C$_{c'}$ or C$_{c''}$), 127.7 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.6 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.5 (aniline C̲H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.4 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.7 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.0 (aniline C̲H, C$_d$, C$_{d'}$ or C$_{d''}$), 105.8 (aromatic C̲H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$, and C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 105.3 (aromatic C̲H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 99.8 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$), 98.5 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$), 98.0 (aromatic C̲H, C$_j$, C$_{j'}$ or C$_{j''}$), 55.3 (C̲H$_3$, C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$), 55.2 (C̲H$_3$, C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$), 55.1 (C̲H$_3$, C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$) **Analysis calculated** (+ 3 CH$_2$Cl$_2$) C 49.79, H 4.18, N 7.74 % **Analysis found** C 50.45, H 4.57, N 7.72 % **ES MS (+)** (CH$_3$OH) m/z 831.22 [MH$^+$] **IR** (cm^{-1}) ν 3084 (w, ArCH), 2938 (w, MeCH), 2837 (w), 1584 (s, CO), 1456 (m), 1283 (m), 1201 (m), 1149 (s), 1058 (m), 828 (w), 764 (m), 682 (m)

8.8 Syntheses of cobalt(III) *tris*-picolinamide complexes (mixed coordination)

The required picolinamide ligand (3 equivalents) in ethanol (10 mL) was added to a solution of cobalt(II) chloride hexahydrate (1 equivalent) in ethanol (10 mL). Triethylamine (3 equivalents) was added and the mixture was heated at 85 °C for 2 hours to give a brown solution. The solvent was removed under reduced pressure and the product purified by column chromatography (methanol/diethyl ether). Product was dissolved in dichloromethane or chloroform, filtered through a sinter and the pure product was precipitated with the addition of hexane.

8.8.1 Synthesis of CoC$_{36}$H$_{27}$N$_6$O$_3$ (2.1b)

Purified by column chromatography (methanol/diethyl ether 9:1) to give a brown solid (0.090 g, 0.138 mmol, 33 %). Recrystallisation by vapour diffusion (chloroform/diethyl ether) at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
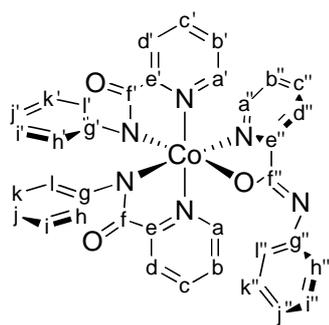
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.0 K) δ 8.99 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.43 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 8.03 (m, 2H, H$_a$, H$_{a'}$ or H$_{a''}$ and H$_c$, H$_{c'}$ or H$_{c''}$), 7.95 (t, 2H, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_c$, H$_{c'}$ or H$_{c''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz), 7.76 (d, 2H, H$_d$, H$_{d'}$ or H$_{d''}$ and H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 7.70 (m, 2H, H$_a$, H$_{a'}$ or H$_{a''}$ and H$_b$, H$_{b'}$ and H$_{b''}$), 7.65 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.2 Hz, ^3J (^{1}H-^{1}H) = 5.6 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.35 (t, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz), 7.10 (m, 3H, H$_b$, H$_{b'}$ or H$_{b''}$, and H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$), 6.98 (m, 8H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_j$, H$_{j'}$ or H$_{j''}$), 6.92 (br. s, 3H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, and H$_j$, H$_{j'}$ or H$_{j''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.1 K) δ 170.1 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 169.3 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$ and C$_f$, C$_{f'}$ or C$_{f''}$), 160.1 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 158.3 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 155.9 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 148.8 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.4 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.3 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 147.2 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 145.5 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 144.7 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 139.4 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.3 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.2 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 128.9 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 128.8 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 128.5 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 127.2 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$ and C$_b$, C$_{b'}$ or C$_{b''}$), 127.1 (aniline $\underline{C}$H, C$_b$,

$C_{b'}$ or $C_{b''}$), 126.5 (aromatic $\underline{C}$H, C_i and C_k, $C_{i'}$ and $C_{k'}$ or $C_{i''}$ and $C_{k''}$), 126.2 (aromatic $\underline{C}$H, C_i and C_k, $C_{i'}$ and $C_{k'}$ or $C_{i''}$ and $C_{k''}$), 126.1 (aromatic $\underline{C}$H, C_i and C_k, $C_{i'}$ and $C_{k'}$ or $C_{i''}$ and $C_{k''}$), 125.5 (aniline $\underline{C}$H, C_d, $C_{d'}$ or $C_{d''}$), 125.4 (aniline $\underline{C}$H, C_d, $C_{d'}$ or $C_{d''}$), 125.3 (aniline $\underline{C}$H, C_d, $C_{d'}$ or $C_{d''}$), 125.1 (aromatic $\underline{C}$H, C_j, $C_{j'}$ or $C_{j''}$), 125.0 (aromatic $\underline{C}$H, C_j, $C_{j'}$ or $C_{j''}$), 124.3 (aromatic $\underline{C}$H, C_j, $C_{j'}$ or $C_{j''}$) **Analysis calculated** (+ 2 H_2O) C 62.97, H 4.55, N 12.24 % **Analysis found** C 63.20, H 4.40, N 12.20 % **ES MS (+)** (CH_3OH) m/z 651.16 [MH$^+$] **IR** (cm^{-1}) ν 3056 (w, ArCH), 1638 (m, CO), 1624 (m, CO), 1600 (m, CO), 1575 (s), 1487 (m), 1384 (m), 1348 (m) 1137 (w), 804 (w), 753 (s), 680 (s), 510 (m)

8.8.2 Synthesis of CoC$_{39}$H$_{33}$N$_6$O$_3$ (2.14b)

Purified by column chromatography (methanol/diethyl ether 7:3) to give a brown solid (0.072 g, 0.104 mmol, 25 %). Recrystallisation by slow evaporation from methanol at room temperature yielded brown crystals suitable for single crystal X-ray diffraction.
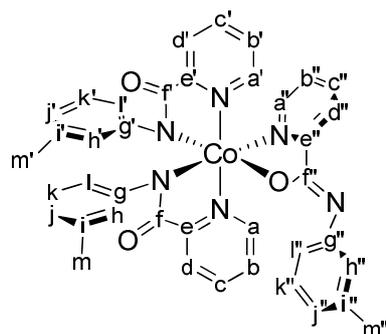
^{1}H NMR (CDCl$_3$, 500.23 MHz, 299.9 K) δ 8.98 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.42 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 8.03 (m, 3H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.93 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.66 (m, 5H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_c$, H$_{c'}$ or H$_{c''}$), 7.24 (t, 1H, H$_k$, H$_{k'}$ or H$_{k''}$, ^3J (^{1}H-^{1}H) = 7.7 Hz), 6.96 (m, 6H, H$_b$, H$_{b'}$ or H$_{b''}$, H$_h$, H$_{h'}$ or H$_{h''}$, H$_h$, H$_{h'}$ or

H$_{h''}$, H$_j$, H$_{j'}$ or H$_{j''}$, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 6.87 (t, 1H, H$_k$, H$_{k'}$ or H$_{k''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 6.77 (m, 3H, H$_h$, H$_{h'}$ or H$_{h''}$, H$_j$, H$_{j'}$ or H$_{j''}$ and H$_k$, H$_{k'}$ or H$_{k''}$), 6.51 (br. s, 2H, H$_l$, H$_{l'}$ or H$_{l''}$ and H$_l$, H$_{l'}$ or H$_{l''}$), 2.39 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 2.10 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 2.06 (br. s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.6 MHz, 299.9 K) δ 170.0 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$ and C$_f$, C$_{f'}$ or C$_{f''}$), 169.2 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 160.2 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.1 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$ and C$_e$, C$_{e'}$ or C$_{e''}$), 148.8 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.4 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.2 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 147.1 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ and C$_{g''}$), 139.3 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.1 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.0 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 138.6 (quaternary aromatic $\underline{C}$, C$_i$, C$_{i'}$ or C$_{i''}$), 138.4 (quaternary aromatic $\underline{C}$, C$_i$, C$_{i'}$ or C$_{i''}$), 138.1 (quaternary aromatic $\underline{C}$, C$_i$, C$_{i'}$ or C$_{i''}$), 128.7 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$ or C$_{h''}$), 128.6 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$ or C$_{h''}$), 128.4 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$ or C$_{h''}$), 127.6 (aromatic $\underline{C}$H, C$_k$, C$_{k'}$ or C$_{k''}$), 127.0 (aromatic $\underline{C}$H, C$_k$, C$_{k'}$ or C$_{k''}$ and C$_k$, C$_{k'}$ or C$_{k''}$), 126.3 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.1 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$ and C$_b$, C$_{b'}$ or C$_{b''}$), 126.0 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.8 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 125.3 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$ and C$_j$, C$_{j'}$ or C$_{j''}$), 125.1 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$), 122.3 (aromatic

CH, C_l, $C_{l'}$ and $C_{l''}$), 21.6 ($\underline{C}H_3$, C_m, $C_{m'}$ or $C_{m''}$), 21.1 ($\underline{C}H_3$, C_m, $C_{m'}$ or $C_{m''}$), 21.0 ($\underline{C}H_3$, C_m, $C_{m'}$ or $C_{m''}$) **Analysis calculated** (+ ½ CH_2Cl_2) C 64.54, H 4.66, N 11.43 % **Analysis found** C 64.22, H 4.46, N 11.41 % **ES MS (+)** (CH_3OH) *m/z* 693.20 [MH^+] **IR** (cm^{-1}) 3398 (br. w, NH), 3032 (w, ArCH), 2917 (w, MeCH), 1621 (m, CO), 1590 (s, CO), 1559 (s, CO), 1480 (m), 1375 (m), 1331 (m), 1261 (m), 1123 (w), 1089 (w), 761 (m), 681 (m)

8.8.3 Synthesis of CoC$_{39}$H$_{33}$N$_6$O$_3$ (2.15b)

Purified by column chromatography (methanol/diethyl ether 4:1) to give a brown solid (0.073 g, 0.105 mmol, 25 %). Recrystallisation by vapour diffusion (chloroform/pentane) at 5 °C yielded brown crystals suitable for single crystal X-ray diffraction.
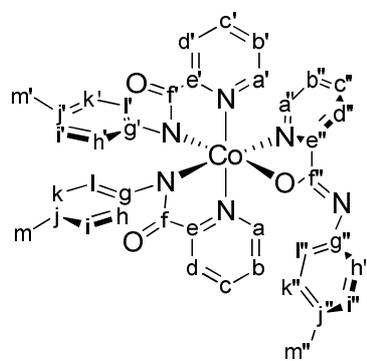
^{1}H NMR (CDCl$_3$, 400.13 MHz, 300.0 K) δ 8.97 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 8.41 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.9 Hz), 7.98 (m, 5H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_b$, H$_{b'}$ or H$_{b''}$, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.70 (m, 4H, H$_b$, H$_{b'}$ or H$_{b''}$, H$_c$, H$_{c'}$ or H$_{c''}$, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.62 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.4 Hz and ^4J (^{1}H-^{1}H) = 1.3 Hz), 7.15 (d, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, ^3J (^{1}H-^{1}H) = 8.1 Hz), 6.96 (m, 6H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$ and, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$), 6.76 (d, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, ^3J (^{1}H-^{1}H) = 7.9 Hz), 6.72 (br. s, 2H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$), 2.35 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 2.07 (s, 6H, H$_m$, H$_{m'}$ or H$_{m''}$ and H$_m$, H$_{m'}$ or H$_{m''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 169.3 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ and C$_{f''}$), 160.2 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 158.4 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.0 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 148.8 (aniline $\underline{C}H$, C$_a$, C$_{a'}$ or C$_{a''}$), 148.3 (aniline $\underline{C}H$, C$_a$, C$_{a'}$ or C$_{a''}$), 148.2 (aniline $\underline{C}H$, C$_a$, C$_{a'}$ or C$_{a''}$), 142.8 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$ and C$_g$, C$_{g'}$ or C$_{g''}$), 142.0 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 139.1 (aniline $\underline{C}H$, C$_c$, C$_{c'}$ or C$_{c''}$), 139.0 (aniline $\underline{C}H$, C$_c$, C$_{c'}$ or C$_{c''}$), 138.9 (aniline $\underline{C}H$, C$_c$, C$_{c'}$ or C$_{c''}$), 134.8 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 134.5 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 134.0 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 129.5 (aromatic $\underline{C}H$, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 129.4 (aromatic $\underline{C}H$, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 129.2 (aromatic $\underline{C}H$, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 126.9 (aniline $\underline{C}H$, C$_b$, C$_{b'}$ or C$_{b''}$ and C$_b$, C$_{b'}$ or C$_{b''}$), 126.7 (aniline $\underline{C}H$, C$_b$, C$_{b'}$ or C$_{b''}$), 126.3 (aniline $\underline{C}H$, C$_d$, C$_{d'}$ or C$_{d''}$), 126.1 (aniline $\underline{C}H$, C$_d$, C$_{d'}$ or C$_{d''}$), 126.0 (aniline $\underline{C}H$, C$_d$, C$_{d'}$ or C$_{d''}$), 125.5 (aromatic $\underline{C}H$, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 125.4 (aromatic $\underline{C}H$, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 125.0 (aromatic $\underline{C}H$, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 21.1 ($\underline{C}H_3$, C$_m$, C$_{m'}$ or C$_{m''}$), 21.0 ($\underline{C}H_3$, C$_m$, C$_{m'}$ or C$_{m''}$ and C$_m$, C$_{m'}$ or C$_{m''}$) **Analysis calculated** (+ $^4/_5$ CH_2Cl_2) C 62.85,

H 4.59, N 11.05 % **Analysis found** C 62.38, H 4.86, N 11.24 % **ES MS (+)** (CH$_3$OH) *m/z* 693.20 [MH$^+$] **IR** (cm^{-1}) ν 3025 (w, ArCH), 2919 (w, MeCH), 1580 (s, CO), 1567 (s, CO), 1505 (m), 1341 (w), 1251 (w), 1096 (w), 806 (m), 757 (m), 680 (m), 510 (s)

8.8.4 Synthesis of CoC$_{42}$H$_{39}$N$_6$O$_3$ (2.16b)

Purified by column chromatography (methanol/diethyl ether 3:2) to give a brown solid (0.106 g, 0.145 mmol, 34 %). Recrystallisation by slow evaporation from methanol at room temperature yielded brown crystals suitable for single crystal X-ray diffraction.
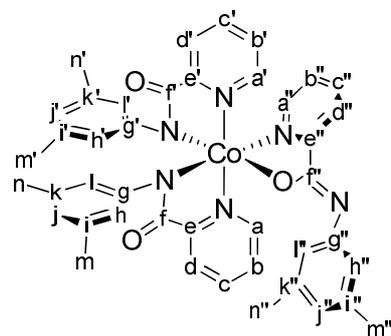
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.0 K) δ 8.98 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.0 Hz), 8.48 (br. s, 1H, H$_d$, H$_{d'}$ or H$_{d''}$), 8.06 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 8.01 (m, 2H, H$_{c'}$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.94 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.70 (m, 2H, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_c$, H$_{c'}$ or H$_{c''}$), 7.64 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.0 Hz, ^3J (^{1}H-^{1}H) = 5.5 Hz and ^4J (^{1}H-^{1}H) = 1.2 Hz), 7.50 (s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or

H$_{h''}$ and H$_{l''}$), 7.15 (br. s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 6.97 (m, 3H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_b$, H$_{b'}$ or H$_{b''}$ and H$_b$, H$_{b'}$ or H$_{b''}$), 6.78 (s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 6.63 (s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.57 (s, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.32 (br. s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 2.36 (s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$), 2.19 (br. s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$), 2.08 (s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.6 MHz, 300.1 K) δ 169.0 (quaternary <u>C</u>-O, C$_f$, C$_{f'}$ and C$_{f''}$), 158.7 (quaternary aniline <u>C</u>, C$_e$, C$_{e'}$ and C$_{e''}$), 148.7 (aniline <u>C</u>H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.4 (aniline <u>C</u>H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.1 (aniline <u>C</u>H, C$_a$, C$_{a'}$ or C$_{a''}$), 144.3 (quaternary aromatic <u>C</u>, C$_g$, C$_{g'}$ and C$_{g''}$), 139.3 (aniline <u>C</u>H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.1 (aniline <u>C</u>H, C$_c$, C$_{c'}$ or C$_{c''}$), 138.9 (aniline <u>C</u>H, C$_c$, C$_{c'}$ or C$_{c''}$), 137.9 (quaternary aromatic <u>C</u>, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$, and C$_{i''}$ and C$_{k''}$), 127.1 (aniline <u>C</u>H, C$_b$, C$_{b'}$ or C$_{b''}$), 127.0 (aniline <u>C</u>H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.9 (aniline <u>C</u>H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.3 (aniline <u>C</u>H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.1 (aniline <u>C</u>H, C$_d$, C$_{d'}$ or C$_{d''}$), 126.0 (aniline <u>C</u>H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.6 (aromatic <u>C</u>H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 125.1 (aromatic <u>C</u>H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 124.6 (aromatic <u>C</u>H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 123.2 (aromatic <u>C</u>H, C$_j$, C$_{j'}$ and C$_{j''}$), 21.6 (<u>C</u>H$_3$, C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$ and C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$), 21.0 (<u>C</u>H$_3$, C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$) **Analysis calculated** (+ ½ CHCl$_3$) C 64.26, H 5.01, N 10.58 % **Analysis found** C 64.30, H 5.50, N 10.80 % **ES MS (+)** (CH$_3$OH) *m/z* 735.25 [MH$^+$] **IR** (cm^{-1}) ν 3066 (w, ArCH), 2914 (w, MeCH), 1620 (m, CO), 1583 (s, CO), 1562 (s, CO), 1467 (m), 1376 (m), 1335 (m), 1260 (w), 1159 (w), 1123 (w), 1028 (w), 840 (m), 762 (m), 682 (m)

8.8.5 Synthesis of CoC$_{39}$H$_{33}$N$_6$O$_6$ (2.17b)

Purified by column chromatography (methanol/diethyl ether 4:1) to give a brown solid (0.055 g, 0.074 mmol, 18 %). Recrystallisation by slow evaporation from methanol at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
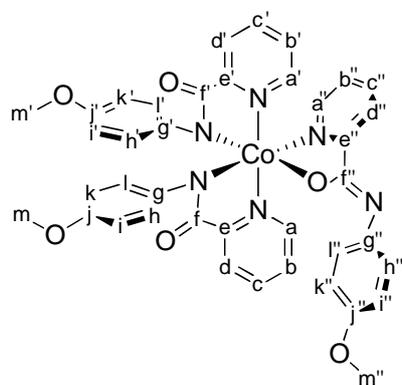
^{1}H NMR (CDCl$_3$, 400.13 MHz, 300.0 K) δ 8.93 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 8.39 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.9 Hz), 8.05 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 7.96 (m, 3H, H$_c$, H$_{c'}$ or H$_{c''}$, H$_d$, H$_{d'}$ or H$_{d''}$ and H$_d$, H$_{d'}$ or H$_{d''}$), 7.82 (m, 2H, H$_a$, H$_{a'}$ or H$_{a''}$ and H$_c$, H$_{c'}$ or H$_{c''}$), 7.72 (m, 2H, H$_b$, H$_{b'}$ or H$_{b''}$ and H$_c$, H$_{c'}$ or H$_{c''}$), 7.60 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.3 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.5 Hz), 6.99 (m, 5H, H$_b$, H$_{b'}$ or H$_{b''}$, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, and H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$), 6.89 (d, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, ^3J (^{1}H-^{1}H) = 9.0 Hz), 6.53 (d, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$, ^3J (^{1}H-^{1}H) = 9.0 Hz), 6.48 (br. s, 4H, H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$, and H$_i$ and H$_k$, H$_{i'}$ and H$_{k'}$ or H$_{i''}$ and H$_{k''}$), 3.82 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 3.65 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$), 3.64 (s, 3H, H$_m$, H$_{m'}$ or H$_{m''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 170.4 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$ and C$_f$, C$_{f'}$ or C$_{f''}$), 169.6 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f''}$), 163.0 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 160.3 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 158.3 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 156.9 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 156.5 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 155.9 (quaternary aromatic $\underline{C}$, C$_j$, C$_{j'}$ or C$_{j''}$), 148.8 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.3 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.2 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 140.7 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 139.2 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.1 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.0 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 138.4 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 137.5 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 127.8 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.8 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.7 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.4 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$, and C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 126.1 (aromatic $\underline{C}$H, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 125.8 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.4 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 125.0 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 114.2 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$, and C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 113.7 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ or C$_{h''}$ and C$_{l''}$), 55.4 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$), 55.3 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$), 55.2 ($\underline{C}$H$_3$, C$_m$, C$_{m'}$ or C$_{m''}$) **Analysis calculated** (+ 1 CH$_2$Cl$_2$) C 60.58, H 4.62, N 10.73 % **Analysis found** C 60.48, H 4.65, N 10.79 % **ES MS (+)** (CH$_3$OH) *m/z* 741.19 [MH$^+$] **IR** (cm^{-1}) ν 3054 (w, ArCH), 2929 (w, MeCH), 2829 (w, MeCH), 1582 (s, CO), 1502 (s), 1338 (m), 1281 (m), 1238 (s), 1168 (m), 1020 (m), 824 (m), 759 (m), 682 (m), 560 (m)

8.8.6 Synthesis of CoC$_{42}$H$_{39}$N$_6$O$_9$ (2.18b)

Purified by column chromatography (methanol/diethyl ether 2:3) to give a brown solid (0.075 g, 0.091 mmol, 22 %). Recrystallisation by slow evaporation from methanol at room temperature yielded orange crystals suitable for single crystal X-ray diffraction.
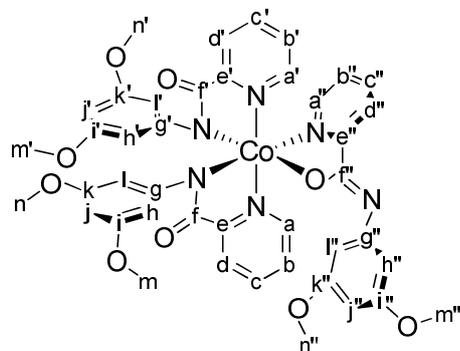
^{1}H NMR (CDCl$_3$, 400.13 MHz, 300.0 K) δ 8.94 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 8.39 (d, 1H, H$_d$, H$_{d'}$ or H$_{d''}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 8.11 (d, 1H, H$_a$, H$_{a'}$ or H$_{a''}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 7.98 (m, 3H, H$_c$, H$_{c'}$ or H$_{c''}$ H$_c$, H$_{c'}$ or H$_{c''}$ and H$_d$, H$_{d'}$ and H$_{d''}$), 7.76 (m, 2H, H$_c$, H$_{c'}$ or H$_{c''}$ and H$_c$, H$_{c'}$ or H$_{c''}$), 7.65 (ddd, 1H, H$_b$, H$_{b'}$ or H$_{b''}$, ^3J (^{1}H-^{1}H) = 7.2 Hz, ^3J (^{1}H-^{1}H) = 5.5 Hz and ^4J (^{1}H-^{1}H) = 1.3

Hz), 7.06 (m, 3H, H$_a$, H$_{a'}$ or H$_{a''}$, H$_b$, H$_{b'}$ or H$_{b''}$ and H$_b$, H$_{b'}$ or H$_{b''}$), 6.88 (m, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.64 (br. s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 6.27 (m, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 6.22 (m, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 6.08 (m, 2H, H$_h$ and H$_l$, H$_{h'}$ and H$_{l'}$ or H$_{h''}$ and H$_{l''}$), 5.98 (br.s, 1H, H$_j$, H$_{j'}$ or H$_{j''}$), 3.76 (s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$), 3.62 (s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$), 3.57 (s, 6H, H$_m$ and H$_n$, H$_{m'}$ and H$_{n'}$ or H$_{m''}$ and H$_{n''}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 169.9 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f'}$), 169.0 (quaternary $\underline{C}$-O, C$_f$, C$_{f'}$ or C$_{f'}$ and C$_f$, C$_{f'}$ or C$_{f'}$), 164.4 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 160.7 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 160.6 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$, C$_{i'}$ and C$_{k'}$ or C$_{i''}$ and C$_{k''}$), 158.4 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$ and C$_e$, C$_{e'}$ or C$_{e''}$), 156.1 (quaternary aniline $\underline{C}$, C$_e$, C$_{e'}$ or C$_{e''}$), 149.0 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.7 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 148.4 (aniline $\underline{C}$H, C$_a$, C$_{a'}$ or C$_{a''}$), 147.3 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$ and C$_g$, C$_{g'}$ or C$_{g''}$), 146.7 (quaternary aromatic $\underline{C}$, C$_g$, C$_{g'}$ or C$_{g''}$), 139.4 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.3 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 139.2 (aniline $\underline{C}$H, C$_c$, C$_{c'}$ or C$_{c''}$), 127.2 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.3 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 126.1 (aniline $\underline{C}$H, C$_b$, C$_{b'}$ or C$_{b''}$), 125.2 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$ and C$_d$, C$_{d'}$ or C$_{d''}$), 125.0 (aniline $\underline{C}$H, C$_d$, C$_{d'}$ or C$_{d''}$), 103.2 (aromatic $\underline{C}$H, C$_h$ and C$_l$, C$_{h'}$ and C$_{l'}$ and C$_{h''}$ and C$_{l''}$), 99.6 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$), 98.2 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$), 97.1 (aromatic $\underline{C}$H, C$_j$, C$_{j'}$ or C$_{j''}$), 55.3 ($\underline{C}$H$_3$, C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$, and C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$), 55.2 ($\underline{C}$H$_3$, C$_m$ and C$_n$, C$_{m'}$ and C$_{n'}$ or C$_{m''}$ and C$_{n''}$) **Analysis calculated** (+ 2 CH$_2$Cl$_2$) C 52.82, H 4.33, N 8.40 % **Analysis found** C 53.14, H 4.25, N 8.74 % **ES MS (+)** (CH$_3$OH) m/z 831.22 [MH$^+$] **IR** (cm^{-1}) ν 3081 (w, ArCH), 2936 (w, MeCH), 2834 (w, MeCH), 1562 (s, CO), 1454 (m), 1330 (m), 1197 (m), 1145 (s), 1047 (m), 760 (m), 681 (m)

8.9 Syntheses of cobalt(II) *bis*-picolinamide thiocyanate complexes

The required picolinamide ligand (2 equivalents) in ethanol (10 mL) was added to a solution of cobalt(II) nitrate hexahydrate (1 equivalent) in distilled water (5 mL) and stirred at 50 °C for 15 min. Potassium thiocyanate (2 equivalents) in distilled water (5 mL) was added dropwise and the mixture was stirred at 50 °C for 2 hours to give a dark pink solution. Slow evaporation of solvent from the reaction mixture yielded pure product.

8.9.1 Synthesis of CoC$_{26}$H$_{20}$N$_6$O$_2$S$_2$ (3.1)

Dark pink crystals suitable for single crystal X-ray diffraction (0.175 g, 0.337 mmol, 73 %).
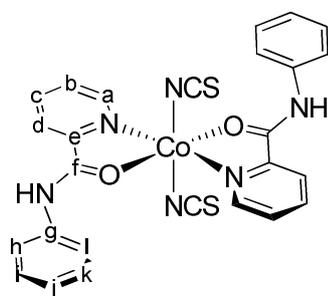
^{1}H NMR (d_6-DMSO, 300.13 MHz, 300.0 K) δ 10.54 (2H, NH), 8.68 (2H, H$_a$), 8.01 (4H, H$_c$ and H$_d$), 7.88 (4H, H$_h$ and H$_l$), 7.62 (2H, H$_b$), 7.26 (4H, H$_i$ and H$_k$), 7.04 (2H, H$_j$) **Analysis calculated** C 54.64, H 3.53, N 14.70 % **Analysis found** C 54.45, H 3.50, N 15.10 % **ES MS (+)** (CH$_3$CN) *m/z* 513.07 [M-NCS$^+$] **IR** (cm^{-1}) ν 3295 (br. w, NH), 3054 (w, ArCH), 2080 (s, CN), 1632 (m, CO), 1605 (m, CO), 1551 (m), 1341 (m), 1261 (m), 1019 (m), 903 (m), 747 (s), 706 (s), 508 (m)

8.9.2 Synthesis of CoC$_{28}$H$_{24}$N$_6$O$_2$S$_2$ (3.2)

Pink crystals suitable for single crystal X-ray diffraction (0.246 g, 0.410 mmol, 98 %).
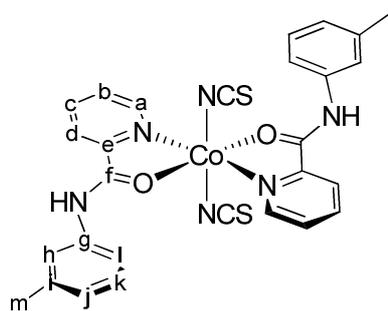
^{1}H NMR (d_6-DMSO, 300.13 MHz, 300.0 K) δ 10.46 (2H, NH), 8.68 (2H, H$_a$), 8.03 (4H, H$_c$ and H$_d$), 7.71 (2H, H$_h$), 7.64 (4H, H$_b$ and H$_j$), 7.16 (2H, H$_k$), 6.89 (2H, H$_l$), 2.24 (6H, H$_m$) **Analysis calculated** C 56.09, H 4.03, N 14.02 % **Analysis found** C 55.97, H 4.02, N 13.95 % **ES MS (+)** (CH$_3$CN) *m/z* 541.10 [M-NCS$^+$] **IR** (cm^{-1}) ν 3255 (br. w, NH), 3067 (w, ArCH), 2918 (w, MeCH), 2083 (s, CN), 1630 (s, CO), 1587 (m, CO), 1551 (s), 1438 (m), 1261 (w), 1020 (w), 781 (m), 682 (m), 645 (m)

8.9.3 Synthesis of CoC$_{30}$H$_{28}$N$_6$O$_2$S$_2$ (3.3)

Dark pink crystals suitable for single crystal X-ray diffraction (0.233 g, 0.372 mmol, 89 %).
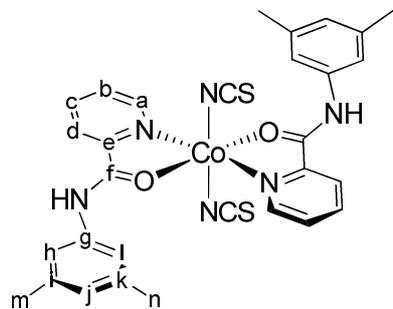
¹H NMR (*d$_6$*-DMSO, 500.23 MHz, 300.1 K) δ 10.27 (2H, NH), 8.60 (2H, H$_a$), 7.96 (4H, H$_c$ and H$_d$), 7.45 (6H, H$_b$, H$_h$ and H$_l$), 6.67 (2H, H$_j$), 2.12 (12H, H$_m$ and H$_n$) **Analysis calculated** C 57.41, H 4.50, N 13.39 % **Analysis found** C 57.50, H 4.60, N 13.60 % **ES MS (+)** (CH$_3$CN) *m/z* 569.13 [M-NCS$^+$] **IR** (cm^{-1}) ν 3263 (br. w, NH), 3099 (w, ArCH), 2912 (w, MeCH). 2098 (s, CN), 1632 (m, CO), 1557 (m, CO), 1433 (m), 1309 (w), 1020 (w), 824 (m), 746 (m), 678 (s)

8.9.4 Synthesis of CoC$_{28}$H$_{24}$N$_6$O$_2$S$_2$ (3.4)

Pink crystals suitable for single crystal X-ray diffraction (0.228 g, 0.382 mmol, 91 %).
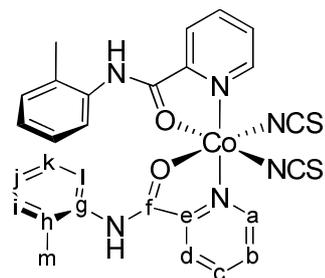
¹H NMR (*d$_6$*-DMSO, 300.13 MHz, 300.0 K) δ 10.21 (2H, NH), 8.70 (2H, H$_a$), 8.12 (2H, H$_d$), 8.03 (2H, H$_c$), 7.82 (2H, H$_l$), 7.64 (2H, H$_b$), 7.22 (4H, H$_i$ and H$_j$), 7.07 (2H, H$_k$), 2.28 (6H, H$_m$) **Analysis calculated** C 56.09, H 4.03, N 14.02 % **Analysis found** C 55.98, H 4.21, N 13.97 % **ES MS (+)** (CH$_3$CN) *m/z* 541.10 [M-NCS$^+$] **IR** (cm^{-1}) ν 3225 (br. w, NH), 3066 (w, ArCH), 2872 (w, MeCH), 2069 (s, CN), 1632 (m, CO), 1589 (m, CO), 1523 (s), 1459 (m), 1304 (w), 997 (w), 917 (w), 747 (s), 692 (m), 595 (w)

8.9.5 Synthesis of CoC$_{28}$H$_{24}$N$_6$O$_2$S$_2$ (3.5)

Pink crystals suitable for single crystal X-ray diffraction (0.174 g, 0.291 mmol, 69 %).
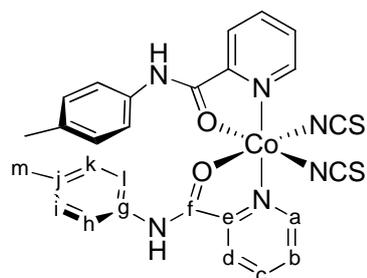
¹H NMR (*d$_6$*-DMSO, 300.13 MHz, 300.0 K) δ 10.47 (2H, NH), 8.68 (2H, H$_a$), 8.03 (4H, H$_c$ and H$_d$), 7.75 (4H, H$_h$ and H$_l$), 7.61 (2H, H$_b$), 7.08 (4H, H$_i$ and H$_k$), 2.20 (6H, H$_m$) **Analysis calculated** (+ 1 H$_2$O) C 54.45, H 4.24, N 13.79 % **Analysis found** C 54.79, H 4.01, N 13.79 % **ES MS (+)** (CH$_3$CN) *m/z* 541.10 [M-NCS$^+$] **IR** (cm^{-1}) ν 3261 (br. w, NH), 3085 (w, ArCH), 2081 (s, CN), 1630 (m, CO), 1586 (m, CO), 1529 (m), 1260 (w), 1020 (w), 808 (m), 751 (m), 687 (m), 596 (m), 509 (s)

8.9.6 Synthesis of CoC$_{28}$H$_{24}$N$_6$O$_2$S$_2$ (3.6)

Purple solid (0.248 g, 0.395 mmol, 94 %).
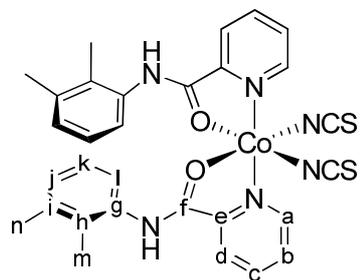
^{1}H NMR (d_6-DMSO, 500.23 MHz, 300.1 K) δ 10.20 (2H, NH), 8.63 (2H, H$_a$), 8.05 (2H, H$_d$), 7.96 (2H, H$_l$), 7.55 (2H, H$_c$), 7.48 (2H, H$_b$), 6.99 (2H, H$_k$), 6.95 (2H, H$_j$), 2.18 (6H, H$_n$), 2.09 (6H, H$_m$) **Analysis calculated** C 57.41, H 4.50, N 13.39 % **Analysis found** C 57.50, H 4.64, N 13.24 % **ES MS (+)** (CH$_3$CN) *m/z* 569.13 [M-NCS$^+$] **IR** (cm^{-1}) v 3264 (br. w, NH), 3068 (w, ArCH), 2915 (w, MeCH), 2070 (s, CN), 1633 (m, CO), 1589 (m, CO), 1531 (s), 1471 (m), 1304 (w), 1264 (w), 1017 (w), 780 (m), 694 (m), 576 (w)

8.9.7 Synthesis of CoC$_{28}$H$_{24}$N$_6$O$_4$S$_2$ (3.7)

Pink crystals suitable for single crystal X-ray diffraction (0.146 g, 0.231 mmol, 55 %).
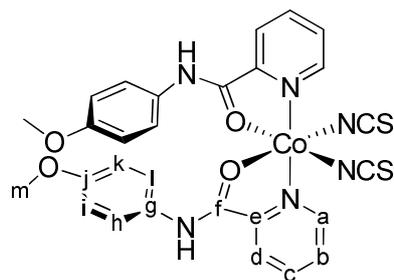
^{1}H NMR (d_6-DMSO, 500.57 MHz, 299.3 K) δ 10.47 (2H, NH), 8.68 (2H, H$_a$), 8.09 (2H, H$_d$), 8.01 (2H, H$_c$), 7.78 (4H, H$_h$ and H$_l$), 7.61 (2H, H$_b$), 6.88 (4H, H$_i$ and H$_k$), 3.49 (6H, H$_m$) **Analysis calculated** C 53.25, H 3.83, N 13.31 % **Analysis found** C 53.33, H 3.70, N 13.24 % **ES MS (+)** (CH$_3$CN) *m/z* 573.09 [M-NCS$^+$] **IR** (cm^{-1}) v 3300 (br. w, NH), 3092 (w, ArCH), 2837 (w, MeCH), 2081 (s, CN), 1626 (m, CO), 1585 (m, CO), 1537 (m), 1508 (m), 1186 (m), 1020 (m), 827 (m), 751 (m), 550 (m), 524 (m)

8.9.8 Synthesis of CoC$_{30}$H$_{28}$N$_6$O$_6$S$_2$ (3.8)

Purple crystals suitable for single crystal X-ray diffraction (0.127 g, 0.184 mmol, 44 %).
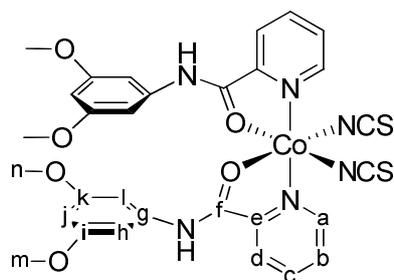
^{1}H NMR (d_6-DMSO, 500.57 MHz, 299.3 K) δ 10.45 (2H, NH), 8.66 (2H, H$_a$), 8.07 (2H, H$_d$), 8.01 (2H, H$_c$), 7.60 (2H, H$_b$), 7.19 (4H, H$_h$ and H$_l$), 6.21 (2H, H$_j$), 3.60 (6H, H$_m$ and H$_n$) **Analysis calculated** C 52.10, H 4.08, N 12.15 % **Analysis found** C 51.97, H 4.09, N 12.01 % **ES MS (+)** (CH$_3$CN) *m/z* 633.11 [M-NCS$^+$] **IR** (cm^{-1}) v 3207 (br. w, NH), 3084 (w, ArCH), 2963 (w, MeCH), 2072 (s, CN), 1612 (m, CO), 1555 (m, CO), 1318 (m), 1158 (s), 1065 (m), 832 (m), 752 (m), 675 (m), 640 (m)

8.10 Synthesis of cobalt(II) *bis*-picolinamide chloride complex

The required picolinamide ligand (2 equivalents) in ethanol (10 mL) was added to a solution of cobalt(II) chloride hexahydrate (1 equivalent) in ethanol (10 mL) and stirred at 50 °C for 2 hours min to give an orange suspension. The solid was filtered, washed with ethanol and dried to yield pure product.

8.10.1 Synthesis of CoC$_{24}$H$_{20}$N$_4$O$_2$Cl$_2$ (3.9)

Orange solid (0.212 g, 0.403 mmol, 96 %). Recrystallisation by slow evaporation from acetonitrile yielded orange crystals suitable for single crystal X-ray diffraction.
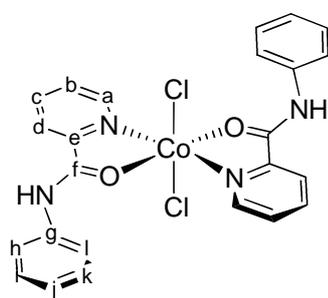
^{1}H NMR (*d$_6$*-DMSO, 300.13 MHz, 300.0 K) δ 10.59 (2H, NH), 8.74 (2H, H$_a$), 8.10 (4H, H$_c$ and H$_d$), 7.91 (4H, H$_h$ and H$_l$), 7.68 (2H, H$_b$), 7.33 (4H, H$_i$ and H$_k$), 7.10 (2H, H$_j$) **Analysis calculated** C 54.77, H 3.83, N 10.65 % **Analysis found** C 54.86, H 3.65, N 10.62 % **ES MS (+)** (CH$_3$CN) *m/z* 490.06 [M-Cl$^+$] **IR** (cm^{-1}) ν 3248 (w, NH), 3092 (w, ArCH), 1630 (m, CO), 1609 (m, CO), 1586 (m), 1558 (m), 1502 (m), 1447 (m), 1347 (m), 1264 (w), 1022 (w), 904 (w), 758 (s), 681 (s), 515 (m)

8.11 Synthesis of cobalt(II) *bis*-picolinamide aqua complex

The required picolinamide ligand (2 equivalents) in ethanol (10 mL) was added to a solution of cobalt(II) chloride hexahydrate (1 equivalent) in distilled water (5 mL) and stirred at 50 °C for 15 min. Potassium iodide (2 equivalents) in distilled water (5 mL) was added dropwise and the mixture was stirred at 50 °C for 2 hours to give an orange solution. Slow evaporation of solvent from the reaction mixture yielded pure product.

8.11.1 Synthesis of CoC$_{24}$H$_{24}$N$_4$O$_4$I$_2$ (3.10)

Orange crystals suitable for single crystal X-ray diffraction (0.304 g, 0.408 mmol, 97 %).
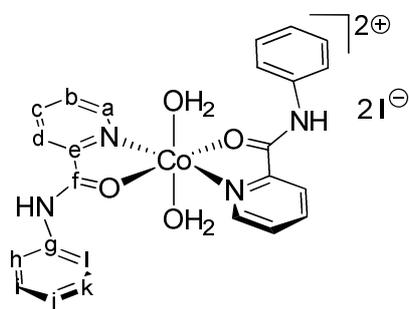
^{1}H NMR (*d$_6$*-DMSO, 300.13 MHz, 300.0 K) δ 10.54 (2H, NH), 8.68 (2H, H$_a$), 8.11 (4H, H$_c$ and H$_d$), 7.86 (4H, H$_h$ and H$_l$), 7.62 (2H, H$_b$), 7.25 (4H, H$_i$ and H$_k$), 7.06 (2H, H$_j$) **Analysis calculated (+ 3 H$_2$O)** C 36.07, H 3.78, N 7.01 % **Analysis found** C 35.85, H 3.18, N 6.66 % **ES MS (+)** (CH$_3$CN) *m/z* 582.00 [M-(2H$_2$O+I)$^+$] **IR** (cm^{-1}) ν 3315 (br. m, OH), 3254 (m, NH), 3084 (w, ArCH), 1626 (m, CO), 1602 (m, CO), 1582 (m), 1547 (m), 1447 (m), 1344 (w), 1262 (w), 1054 (w), 899 (w), 765 (s), 688 (s), 585 (s)

8.12 Synthesis of cobalt(III) *bis*-picolinamide β-diketonate complexes

Complex **4.1** was synthesised and partially characterised by Mr. Bilal Saloo (University of Leeds). Crystals suitable for single crystal X-ray diffraction of complexes **4.1**, **4.3**, **4.8** and **4.9** were provided by Mr. Bilal Saloo (University of Leeds).

The required picolinamide ligand (2 equivalents) and the required β-diketonate ligand (1 equivalent) in ethanol (10 mL) was added to a solution of cobalt(II) chloride hydrate (1 equivalent) in distilled water (5 mL). Hydrogen peroxide (1 equivalent) was added dropwise and the mixture was heated at 85 °C for 18 hours to give a brown solution. The solvent was removed under reduced pressure and the product purified by column chromatography (ethyl acetate/hexane) to give pure product.

8.12.1 Synthesis of CoC₃₃H₂₁N₄O₄F₁₂ (4.1)

Purified by column chromatography (ethyl acetate/hexane 7:3) to give a green solid (0.095 g, 0.115 mmol, 27 %). Recrystallisation by vapour diffusion (dichloromethane/hexane) at room temperature yielded green crystals suitable for single crystal X-ray diffraction.
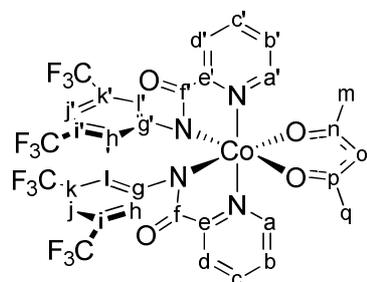
¹H NMR (CDCl₃, 500.23 MHz, 300.0 K) δ 8.14 (d, 2H, H_d and $H_{d'}$, ^3J (^1H-^1H) = 7.5 Hz), 7.92 (td, 2H, H_c and $H_{c'}$, ^3J (^1H-^1H) = 7.7 Hz and ^3J (^1H-^1H) = 1.4 Hz), 7.67 (d, 2H, H_a and $H_{a'}$, ^3J (^1H-^1H) = 5.5 Hz), 7.50 (s, 2H, H_j and $H_{j'}$), 7.45 (br. s, 4H, H_h, $H_{h'}$, H_l and $H_{l'}$), 7.19 (ddd, 2H, H_b and $H_{b'}$, ^3J (^1H-^1H) = 7.0 Hz, ^3J (^1H-^1H) = 5.8 Hz and ^4J (^1H-^1H) = 1.1 Hz), 5.43 (s, 1H, H_o), 2.06 (s, 6H, H_m and H_q) **¹³C {¹H} NMR** (CDCl₃, 125.8 MHz, 300.1 K) δ 190.4 (quaternary diketonate $\underline{C}$, C_n and C_p), 169.3 (quaternary $\underline{C}$-O, C_f and $C_{f'}$), 156.7 (quaternary aniline $\underline{C}$, C_e and $C_{e'}$), 148.5 (quaternary aromatic $\underline{C}$, C_i, $C_{i'}$, C_k and $C_{k'}$), 148.0 (aniline $\underline{C}$H, C_a and $C_{a'}$), 140.0 (aniline $\underline{C}$H, C_c and $C_{c'}$), 131.8 ($\underline{C}$F₃), 128.3 (aromatic $\underline{C}$H, C_h, $C_{h'}$, C_l and $C_{l'}$), 126.5 (aniline $\underline{C}$H, C_b and $C_{b'}$), 125.9 (aniline $\underline{C}$H, C_d and $C_{d'}$), 124.0 (quaternary aromatic $\underline{C}$, C_g and $C_{g'}$), 121.8 (aromatic $\underline{C}$H, C_j or $C_{j'}$), 119.6 (aromatic $\underline{C}$H, C_j or $C_{j'}$), 97.9 (diketonate $\underline{C}$H, C_o), 27.3 ($\underline{C}$H₃, C_m and C_q) **Analysis calculated** C 48.07, H 2.57, N 6.80 % **Analysis found** C 47.92, H 2.43, N 6.78 % **ES MS (+)** (CH₃OH) *m/z* 825.07 [MH⁺] **IR** (cm⁻¹) ν 3090 (w, ArCH), 2962 (w, MeCH), 1637 (m, CO), 1604 (m, CO), 1570 (m, CO), 1518 (m), 1376 (m), 1274 (s), 1056 (s), 919 (m), 770 (m), 681 (m), 617 (w)

8.12.2 Synthesis of CoC₄₃H₂₅N₄O₄F₁₂ (4.2)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a green solid (0.088 g, 0.093 mmol, 22 %). Recrystallisation by slow evaporation from methanol at room temperature yielded green crystals suitable for single crystal X-ray diffraction.
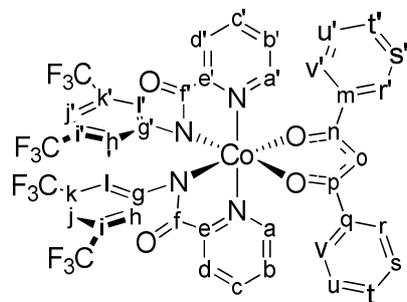
¹H NMR (CDCl₃, 500.23 MHz, 300.2 K) δ 8.20 (d, 2H, H_d and $H_{d'}$, ^3J (^1H-^1H) = 7.7 Hz), 7.90 (m, 6H, H_c, $H_{c'}$, H_j, $H_{j'}$, H_t and $H_{t'}$), 7.81 (d, 2H, H_a and $H_{a'}$, ^3J (^1H-^1H) = 5.5 Hz), 7.59 (m, 6H, H_h, $H_{h'}$, H_l, $H_{l'}$, H_r and $H_{r'}$ or H_v and $H_{v'}$), 7.50 (m, 6H, H_r and $H_{r'}$ or H_v and $H_{v'}$, H_s, $H_{s'}$, H_u and $H_{u'}$), 7.13 (ddd, 2H, H_b and $H_{b'}$, ^3J (^1H-^1H) = 7.5 Hz, ^3J (^1H-^1H) = 5.7 Hz and ^4J (^1H-^1H) = 1.3 Hz), 6.79 (s, 1H, H_o) **¹³C {¹H} NMR** (CDCl₃, 125.8 MHz, 300.1 K) δ 184.0 (quaternary diketonate $\underline{C}$, C_n and C_p), 169.5 (quaternary $\underline{C}$-O, C_f and $C_{f'}$), 156.6 (quaternary aniline $\underline{C}$, C_e or $C_{e'}$), 148.6 (quaternary aromatic $\underline{C}$, C_i, $C_{i'}$, C_k and $C_{k'}$), 147.9 (aniline $\underline{C}$H, C_a and

C$_{a'}$), 140.1 (aniline $\underline{C}$H, C$_c$ and C$_{c'}$), 136.5 (aromatic $\underline{C}$H, C$_r$ and C$_v$), 132.8 (aromatic $\underline{C}$H, C$_t$), 131.9 ($\underline{C}$F$_3$), 128.9 (aromatic $\underline{C}$H, C$_u$ and C$_s$), 127.1 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$, C$_l$ and C$_{l'}$), 126.7 (aniline $\underline{C}$H, C$_b$ and C$_{b'}$), 126.2 (aniline $\underline{C}$H, C$_d$ and C$_{d'}$), 123.9 (quaternary aromatic $\underline{C}$, C$_g$), 121.8 (quaternary aromatic $\underline{C}$, C$_m$ and C$_q$), 118.5 (aromatic $\underline{C}$H, C$_j$ and C$_{j'}$), 91.4 (diketonate $\underline{C}$H, C$_o$) **Analysis calculated** C 54.44, H 2.66, N 5.91 % **Analysis found** C 54.31, H 2.55, N 5.95 % **ES MS (+)** (CH$_3$OH) *m/z* 949.11 [MH$^+$] **IR** (cm^{-1}) v 3086 (w, ArCH), 1639 (m, CO), 1600 (m, CO), 1515 (m), 1373 (m), 1277 (s), 1119 (s), 978 (w), 878 (w), 761 (w), 684 (m). 616 (w)

8.12.3 Synthesis of CoC$_{38}$H$_{23}$N$_4$O$_4$F$_{12}$ (4.3)

Purified by column chromatography (ethyl acetate/hexane 3:7) to give a green solid (0.043 g, 0.048 mmol, 11 %). Recrystallisation by vapour diffusion (dichloromethane/hexane) at room temperature yielded green crystals suitable for single crystal X-ray diffraction.
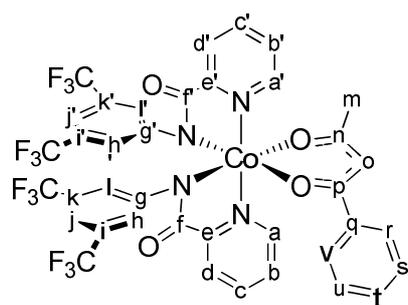
^{1}H NMR (CDCl$_3$, 400.13 MHz, 300.0 K) δ 8.19 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz), 8.15 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.93 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz and ^3J (^{1}H-^{1}H) = 1.2 Hz), 7.90 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz and ^3J (^{1}H-^{1}H) = 1.2 Hz), 7.82 (s, 1H, H$_j$ or H$_{j'}$), 7.80 (s, 1H, H$_j$ or H$_{j'}$), 7.75 (d, 1H, H$_a$ or H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 7.73 (d, 1H, H$_a$ or H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 7.55 (m, 3H, H$_r$, H$_t$ and H$_v$), 7.52 (s, 2H, H$_h$ and H$_l$ or H$_{h'}$ and H$_{l'}$), 7.50 (s, 2H, H$_h$ and H$_l$ or H$_{h'}$ and H$_{l'}$), 7.44 (t, 2H, H$_s$ and H$_u$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.18 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz, ^3J (^{1}H-^{1}H) = 5.9 Hz and ^4J (^{1}H-^{1}H) = 1.5 Hz), 7.14 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.4 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.5 Hz), 6.10 (s, 1H, H$_o$), 2.21 (s, 3H, H$_m$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 192.0 (quaternary diketonate $\underline{C}$, C$_n$), 182.4 (quaternary diketonate $\underline{C}$, C$_p$), 169.4 (quaternary $\underline{C}$-O, C$_f$ and C$_{f'}$), 156.7 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 156.6 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 148.6 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.5 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.0 (aniline $\underline{C}$H, C$_a$ or C$_{a'}$), 147.9 (aniline $\underline{C}$H, C$_a$ or C$_{a'}$), 140.1 (aniline $\underline{C}$H, C$_c$ and C$_{c'}$), 135.9 (aromatic $\underline{C}$H, C$_r$ and C$_v$), 132.7 (aromatic $\underline{C}$H, C$_t$), 131.9 ($\underline{C}$F$_3$), 128.8 (aromatic $\underline{C}$H, C$_u$ and C$_s$), 127.0 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$, C$_l$ and C$_{l'}$), 126.7 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 126.6 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 126.1 (aniline $\underline{C}$H, C$_d$ and C$_{d'}$), 124.2 (quaternary aromatic $\underline{C}$, C$_g$), 121.5 (quaternary aromatic $\underline{C}$, C$_q$), 118.4 (aromatic $\underline{C}$H, C$_j$ and C$_{j'}$), 94.5 (diketonate $\underline{C}$H, C$_o$), 28.1 ($\underline{C}$H$_3$, C$_m$) **Analysis calculated** C 51.48, H 2.61, N 6.32 % **Analysis found** C 51.35, H 2.58, N 6.23 % **ES MS (+)** (CH$_3$OH) *m/z* 887.10 [MH$^+$] **IR** (cm^{-1}) v 3079 (w, ArCH), 1634 (m, CO), 1601 (m, CO), 1551 (m, CO), 1512 (m), 1373 (s), 1273 (s), 1122 (s), 975 (m), 763 (m), 685 (s), 608 (w)

8.12.4 Synthesis of CoC$_{38}$H$_{22}$N$_4$O$_4$F$_{13}$ (4.4)

Purified by column chromatography (ethyl acetate/hexane 3:7) to give a green solid (0.061 g, 0.068 mmol, 16 %). Recrystallisation by slow evaporation from methanol at room temperature yielded green crystals suitable for single crystal X-ray diffraction.
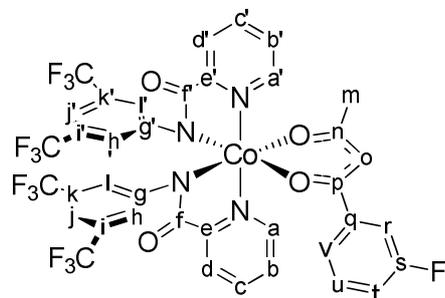
^{1}H NMR (CDCl$_3$, 400.13 MHz, 300.0 K) δ 8.19 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 0.8 Hz), 8.14 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 0.9 Hz), 7.94 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.4 Hz), 7.91 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.4 Hz), 7.82 (m, 2H, H$_r$ and H$_t$), 7.75 (d, 1H, H$_a$ or H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.1 Hz), 7.69 (d, 1H, H$_a$ or H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.1 Hz), 7.51 (m, 6H, H$_h$, H$_{h'}$, H$_j$, H$_{j'}$, H$_l$ and H$_{l'}$), 7.19 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.6 Hz), 7.14 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.4 Hz), 6.04 (s, 1H, H$_o$), 2.20 (s, 3H, H$_m$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 192.2 (quaternary diketonate $\underline{C}$, C$_n$), 181.0 (quaternary diketonate $\underline{C}$, C$_p$), 169.4 (quaternary $\underline{C}$-O, C$_f$ or C$_{f'}$), 156.7 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 156.6 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 148.5 (quaternary aromatic $\underline{C}$, C$_i$, C$_{i'}$, C$_k$ and C$_{k'}$), 147.9 (aniline $\underline{C}$H, C$_a$ or C$_{a'}$), 147.8 (aniline $\underline{C}$H, C$_a$ or C$_{a'}$), 140.1 (aniline $\underline{C}$H, C$_c$ and C$_{c'}$), 132.3 ($\underline{C}$F$_3$), 131.6 (aromatic $\underline{C}$H, C$_u$ and C$_v$), 129.4 (aromatic $\underline{C}$H, C$_r$ and C$_t$), 128.3 (aromatic $\underline{C}$H, C$_h$ and C$_l$, or C$_{h'}$ and C$_{l'}$), 128.2 (aromatic $\underline{C}$H, C$_h$ and C$_l$, or C$_{h'}$ and C$_{l'}$), 126.7 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 126.6 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 126.1 (aniline $\underline{C}$H, C$_d$ or C$_{d'}$), 126.0 (aniline $\underline{C}$H, C$_d$ or C$_{d'}$), 124.2 (quaternary aromatic $\underline{C}$, C$_g$), 121.5 (quaternary aromatic $\underline{C}$, C$_q$), 118.5 (aromatic $\underline{C}$H, C$_s$), 116.0 (aromatic $\underline{C}$H, C$_j$ or C$_{j'}$), 115.8 (aromatic $\underline{C}$H, C$_j$ or C$_{j'}$), 94.3 (diketonate $\underline{C}$H, C$_o$), 28.1 ($\underline{C}$H$_3$, C$_m$) **Analysis calculated** C 50.46, H 2.45, N 6.19 % **Analysis found** C 50.49, H 2.54, N 6.08 % **ES MS (+)** (CH$_3$OH) *m/z* 905.08 [MH$^+$] **IR** (cm^{-1}) ν 3098 (w, ArCH), 2962 (w, MeCH), 1637 (m, CO), 1602 (m, CO), 1498 (m), 1391 (m), 1273 (s), 1118 (s), 977 (m), 846 (w), 764 (w), 686 (m), 524 (w)

8.12.5 Synthesis of CoC₃₈H₂₂N₄O₄F₁₃ (4.5)

8.12.5 Synthesis of $CoC_{38}H_{22}N_4O_4F_{13}$ (4.5)

Purified by column chromatography (ethyl acetate/hexane 3:7) to give a green solid (0.077 g, 0.085 mmol, 20 %).
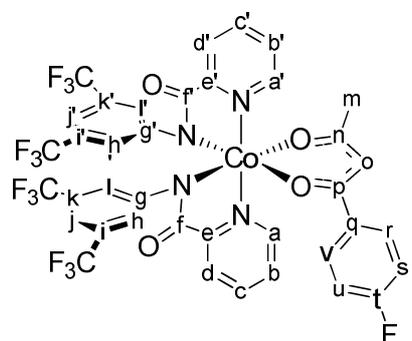
¹H NMR (CDCl₃, 400.13 MHz, 300.0 K) δ 8.16 (overlapping d, 2H, H$_d$ and H$_{d'}$), 7.92 (overlapping td, 1H, H$_c$ or H$_{c'}$), 7.76 (m, 3H, H$_a$, H$_{a'}$ and H$_r$ or H$_v$), 7.51 (m, 6H, H$_h$, H$_{h'}$, H$_j$, H$_{j'}$, H$_l$ and H$_{l'}$), 7.32 (m, 2H, H$_r$ or H$_v$ and H$_s$ or H$_u$), 7.14 (m, 3H, H$_b$, H$_{b'}$ and H$_s$ or H$_u$), 6.08 (s, 1H, H$_o$), 2.20 (s, 3H, H$_m$) **¹³C {¹H} NMR** (CDCl₃, 100.6 MHz, 300.0 K) δ 192.1 (quaternary diketonate $\underline{C}$, C$_n$), 182.1 (quaternary diketonate $\underline{C}$, C$_p$), 169.3 (quaternary $\underline{C}$-O, C$_f$ and C$_{f'}$), 159.9 (aromatic $\underline{C}H$, C$_t$), 156.6 (quaternary aniline $\underline{C}$, C$_e$ and C$_{e'}$), 148.6 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.5 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 147.9 (aniline $\underline{C}H$, C$_a$ or C$_{a'}$), 147.8 (aniline $\underline{C}H$, C$_a$ or C$_{a'}$), 140.1 (aniline $\underline{C}H$, C$_c$ or C$_{c'}$), 140.0 (aniline $\underline{C}H$, C$_c$ or C$_{c'}$), 137.3 (aromatic $\underline{C}H$, C$_r$ and C$_v$), 132.0 ($\underline{C}F_3$), 129.8 (aromatic $\underline{C}H$, C$_u$ and C$_s$), 129.4 (aromatic $\underline{C}H$, C$_h$, C$_{h'}$, C$_l$ and C$_{l'}$), 126.7 (aniline $\underline{C}H$, C$_b$ or C$_{b'}$), 126.6 (aniline $\underline{C}H$, C$_b$ or C$_{b'}$), 126.1 (aniline $\underline{C}H$, C$_d$ or C$_{d'}$), 126.0 (aniline $\underline{C}H$, C$_d$ or C$_{d'}$), 124.2 (quaternary aromatic $\underline{C}$, C$_g$), 121.5 (quaternary aromatic $\underline{C}$, C$_q$), 118.3 (aromatic $\underline{C}H$, C$_j$ and C$_{j'}$), 94.4 (diketonate $\underline{C}H$, C$_o$), 28.1 ($\underline{C}H_3$, C$_m$) **Analysis calculated** C 50.46, H 2.45, N 6.19 % **Analysis found** C 50.58, H 2.53, N 6.24 % **ES MS (+)** (CH₃OH) *m/z* 905.08 [MH⁺] **IR** (cm⁻¹) ν 3080 (w, ArCH), 2920 (w, MeCH), 1636 (m, CO), 1600 (m, CO), 1551 (m, CO), 1373 (m), 1273 (s), 1122 (s), 974 (m), 877 (w), 844 (w), 748 (w), 685 (m)

8.12.6 Synthesis of CoC₃₉H₂₂N₄O₄F₁₅ (4.6)

Purified by column chromatography (ethyl acetate/hexane 3:7) to give a green solid (0.058 g, 0.060 mmol, 14 %). Recrystallisation by slow evaporation from methanol at room temperature yielded green crystals suitable for single crystal X-ray diffraction.
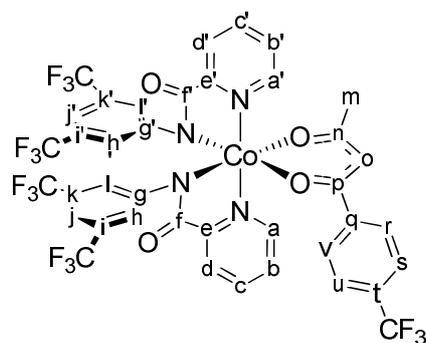
¹H NMR (CDCl₃, 500.23 MHz, 300.2 K) δ 8.20 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.16 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.94 (overlapping td, 2H, H$_c$ and H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz and ^3J (^{1}H-^{1}H) = 1.2 Hz), 7.89 (s, 1H, H$_j$ or H$_{j'}$), 7.87 (s, 1H, H$_j$ or H$_{j'}$), 7.74 (d, 1H, H$_a$ or H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 7.69 (m, 3H, H$_a$ or H$_{a'}$, H$_s$ and H$_u$), 7.52 (m, 6H, H$_h$, H$_{h'}$, H$_l$, H$_{l'}$, H$_r$ and H$_v$), 7.20 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.4 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.4 Hz), 7.16 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H)

= 7.4 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.4 Hz), 6.12 (s, 1H, H$_o$), 2.24 (s, 3H, H$_m$) ^{13}C {^{1}H} NMR (CDCl$_3$, 125.8 MHz, 300.1 K) δ 193.6 (quaternary diketonate $\underline{C}$, C$_n$), 180.6 (quaternary diketonate $\underline{C}$, C$_p$), 169.3 (quaternary $\underline{C}$-O, C$_f$ and C$_{f'}$), 156.6 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 156.5 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$),148.5 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.4 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 147.8 (aniline $\underline{C}$H, C$_a$ or C$_{a'}$), 147.7 (aniline $\underline{C}$H, C$_a$ or C$_{a'}$), 140.2 (aniline $\underline{C}$H, C$_c$ and C$_{c'}$), 139.0 (aromatic $\underline{C}$H, C$_t$), 134.0 ($\underline{C}$F$_3$), 132.9 (diketonate $\underline{C}$F$_3$), 128.3 (aromatic $\underline{C}$H, C$_r$ and C$_v$), 127.3 (aromatic $\underline{C}$H, C$_u$ and C$_s$), 126.8 (aromatic $\underline{C}$H, C$_h$ and C$_l$ or C$_{h'}$ and C$_{l'}$), 126.7 (aromatic $\underline{C}$H, C$_h$ and C$_l$ or C$_{h'}$ and C$_{l'}$), 126.2 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 126.1 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 125.8 (aniline $\underline{C}$H, C$_d$ or C$_{d'}$), 125.7 (aniline $\underline{C}$H, C$_d$ or C$_{d'}$), 123.9 (quaternary aromatic $\underline{C}$, C$_g$), 121.8 (quaternary aromatic $\underline{C}$, C$_q$), 118.6 (aromatic $\underline{C}$H, C$_j$ and C$_{j'}$), 95.2 (diketonate $\underline{C}$H, C$_o$), 28.4 ($\underline{C}$H$_3$, C$_m$) **Analysis calculated** C 49.07, H 2.32, N 5.87 % **Analysis found** C 49.15, H 2.17, N 5.90 % **ES MS (+)** (CH$_3$OH) *m/z* 955.08 [MH$^+$] **IR** (cm^{-1}) v 3087 (w, ArCH), 1637 (m, CO), 1603 (m, CO), 1558 (m, CO), 1553 (m), 1390 (m), 1272 (s), 1115 (s), 976 (m), 775 (m), 686 (m), 632 (w)

8.12.7 Synthesis of CoC$_{39}$H$_{25}$N$_4$O$_4$F$_{12}$ (4.7)

Purified by column chromatography (ethyl acetate/hexane 3:7) to give a green solid (0.052 g, 0.047 mmol, 14 %).
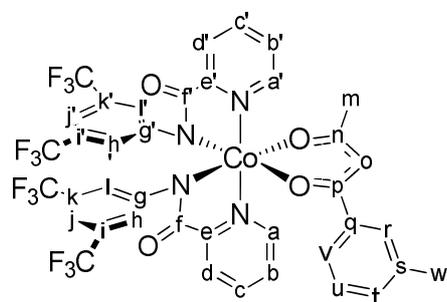
^{1}**H NMR** (CDCl$_3$, 400.13 MHz, 300.0 K) δ 8.18 (dd, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 0.8 Hz), 8.14 (dd, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz and ^3J (^{1}H-^{1}H) = 0.9 Hz), 7.93 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz and ^3J (^{1}H-^{1}H) = 1.3 Hz), 7.90 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.3 Hz), 7.74 (m, 2H, H$_a$ and H$_{a'}$), 7.64 (s, 1H, H$_j$ or H$_{j'}$), 7.55 (m, 7H, H$_j$ or H$_{j'}$, H$_h$, H$_{h'}$, H$_l$, H$_{l'}$, H$_r$ and H$_t$), 7.33 (m, 2H, H$_u$ and H$_v$), 7.16 (overlapping ddd, 2H, H$_b$ and H$_{b'}$, ^4J (^{1}H-^{1}H) = 1.5 Hz), 6.10 (s, 1H, H$_o$), 2.42 (s, 3H, H$_w$), 2.21 (s, 3H, H$_m$) ^{13}**C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 191.7 (quaternary diketonate $\underline{C}$, C$_n$), 182.6 (quaternary diketonate $\underline{C}$, C$_p$), 169.4 (quaternary $\underline{C}$-O, C$_f$ or C$_{f'}$), 169.3 (quaternary $\underline{C}$-O, C$_f$ or C$_{f'}$), 156.7 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 156.6 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 148.6 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.5 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.0 (aniline $\underline{C}$H, C$_a$ or C$_{a'}$), 147.9 (aniline $\underline{C}$H, C$_a$ or C$_{a'}$), 140.0 (aniline $\underline{C}$H, C$_c$ and C$_{c'}$), 138.8 (aromatic $\underline{C}$H, C$_s$), 133.5 (aromatic $\underline{C}$H, C$_r$ and C$_t$), 132.0 (aromatic $\underline{C}$H, C$_u$), 131.9 ($\underline{C}$F$_3$), 128.6 (aromatic $\underline{C}$H, C$_v$), 127.6 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$, C$_l$ and C$_{l'}$), 126.6 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 126.5 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 126.0 (aniline $\underline{C}$H, C$_d$ and C$_{d'}$), 124.3 (quaternary aromatic $\underline{C}$, C$_g$), 121.5 (quaternary aromatic $\underline{C}$, C$_q$), 118.4 (aromatic $\underline{C}$H, C$_j$ and C$_{j'}$), 94.5

(diketonate C̲H, C$_o$), 28.1 (C̲H$_3$, C$_m$), 21.3 (C̲H$_3$, C$_w$) **Analysis calculated** C 52.01, H 2.80, N 6.22 % **Analysis found** C 51.85, H 2.63, N 6.13 % **ES MS (+)** (CH$_3$OH) *m/z* 901.10 [MH$^+$] **IR** (cm^{-1}) ν 3081 (w, ArCH), 2923 (w, MeCH), 1636 (m, CO), 1601 (m, CO), 1551 (m, CO), 1515 (m), 1371 (m), 1273 (s), 1125 (s), 973 (w), 877 (w), 764 (w), 685 (m), 608 (m)

8.12.8 Synthesis of CoC$_{39}$H$_{25}$N$_4$O$_4$F$_{12}$ (4.8)

Purified by column chromatography (ethyl acetate/hexane 3:7) to give a green solid (0.102 g, 0.113 mmol, 27 %). Recrystallisation by vapour diffusion (dichloromethane/hexane) at room temperature yielded green crystals suitable for single crystal X-ray diffraction.
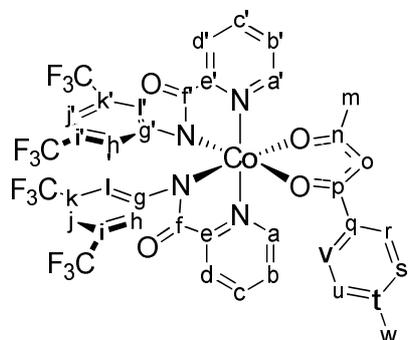
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.0 K) δ 8.16 (overlapping d, 2H, H$_d$ and H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.8 Hz), 7.91 (overlapping td, 2H, H$_c$ and H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.3 Hz), 7.73 (m, 4H, H$_a$, H$_{a'}$, H$_j$ and H$_{j'}$), 7.52 (m, 6H, H$_h$, H$_{h'}$, H$_l$, H$_{l'}$, H$_r$ and H$_v$), 7.24 (m, 2H, H$_u$ and H$_s$), 7.15 (overlapping ddd, 2H, H$_b$ and H$_{b'}$, ^4J (^{1}H-^{1}H) = 1.3 Hz), 6.08 (s, 1H, H$_o$), 2.42 (s, 3H, H$_w$), 2.19 (s, 3H, H$_m$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.2 K) δ 191.4 (quaternary diketonate C̲, C$_n$), 182.3 (quaternary diketonate C̲, C$_p$), 169.3 (quaternary C̲-O, C$_f$ and C$_{f'}$), 156.7 (quaternary aniline C̲, C$_e$ and C$_{e'}$), 148.6 (quaternary aromatic C̲, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.5 (quaternary aromatic C̲, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.0 (aniline C̲H, C$_a$ or C$_{a'}$), 147.9 (aniline C̲H, C$_a$ or C$_{a'}$), 143.7 (aromatic C̲H, C$_t$), 140.0 (aniline C̲H, C$_c$ and C$_{c'}$), 133.2 (aromatic C̲H, C$_r$ and C$_v$), 131.9 (C̲F$_3$), 129.5 (aromatic C̲H, C$_u$ and C$_s$), 127.1 (aromatic C̲H, C$_h$, C$_{h'}$, C$_l$ and C$_{l'}$), 126.6 (aniline C̲H, C$_b$ or C$_{b'}$), 126.5 (aniline C̲H, C$_b$ or C$_{b'}$), 126.0 (aniline C̲H, C$_d$ and C$_{d'}$), 124.0 (quaternary aromatic C̲, C$_g$), 121.8 (quaternary aromatic C̲, C$_q$), 118.4 (aromatic C̲H, C$_j$ and C$_{j'}$), 94.1 (diketonate C̲H, C$_o$), 28.0 (C̲H$_3$, C$_m$), 21.5 (C̲H$_3$, C$_w$) **Analysis calculated** C 52.01, H 2.80, N 6.22 % **Analysis found** C 51.99, H 2.91, N 6.14 % **ES MS (+)** (CH$_3$OH) *m/z* 901.10 [MH$^+$] **IR** (cm^{-1}) ν 3083 (w, ArCH), 2965 (w, MeCH), 1635 (m, CO), 1602 (m, CO), 1519 (m), 1499 (m), 1372 (m), 1273 (s), 1121 (s), 973 (m), 763 (m), 686 (m),608 (w)

8.12.9 Synthesis of CoC$_{39}$H$_{25}$N$_4$O$_5$F$_{12}$ (4.9)

Purified by column chromatography (ethyl acetate/hexane 3:7) to give a green solid (0.102 g, 0.111 mmol, 26 %). Recrystallisation by vapour diffusion (dichloromethane/hexane) at room temperature yielded green crystals suitable for single crystal X-ray diffraction.
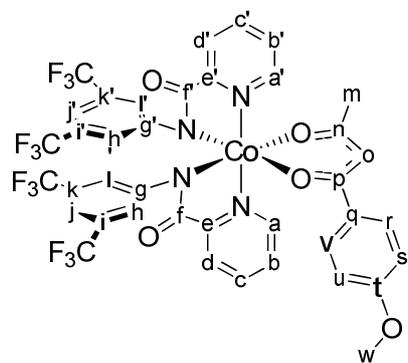
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.1 K) δ 8.17 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.2 Hz), 8.14 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 7.91 (overlapping td, 2H, H$_c$ and H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz and ^3J (^{1}H-^{1}H) = 1.4 Hz), 7.81 (s, 1H, H$_j$ or H$_{j'}$), 7.79 (s, 1H, H$_j$ or H$_{j'}$), 7.75 (d, 1H, H$_a$ or H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 7.71 (d, 1H, H$_a$ or H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 7.52 (m, 6H, H$_h$, H$_{h'}$, H$_l$, H$_{l'}$, H$_r$ and H$_v$), 7.15 (overlapping ddd, 2H, H$_b$ and H$_{b'}$, ^4J (^{1}H-^{1}H) = 1.3 Hz), 6.91 (m, 2H, H$_u$ and H$_s$), 6.03 (s, 1H, H$_o$), 3.88 (s, 3H, H$_w$), 2.18 (s, 3H, H$_m$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.1 K) δ 190.6 (quaternary diketonate C, C$_n$), 181.6 (quaternary diketonate C, C$_p$), 169.4 (quaternary C-O, C$_f$ and C$_{f'}$), 163.4 (aromatic CH, C$_t$), 156.8 (quaternary aniline C, C$_e$ or C$_{e'}$), 156.7 (quaternary aniline C, C$_e$ or C$_{e'}$),148.6 (quaternary aromatic C, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.5 (quaternary aromatic C, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.0 (aniline CH, C$_a$ or C$_{a'}$), 147.9 (aniline CH, C$_a$ or C$_{a'}$), 140.0 (aniline CH, C$_c$ and C$_{c'}$), 132.0 (aromatic CH, C$_r$ and C$_v$), 131.7 (CF$_3$), 129.2 (aromatic CH, C$_u$ and C$_s$), 128.4 (aromatic CH, C$_h$ and C$_l$ or C$_{h'}$ and C$_{l'}$), 128.3 (aromatic CH, C$_h$ and C$_l$ or C$_{h'}$ and C$_{l'}$), 126.6 (aniline CH, C$_b$ or C$_{b'}$), 126.5 (aniline CH, C$_b$ or C$_{b'}$), 126.0 (aniline CH, C$_d$ and C$_{d'}$), 124.0 (quaternary aromatic C, C$_g$), 121.8 (quaternary aromatic C, C$_q$), 118.3 (aromatic CH, C$_j$ and C$_{j'}$), 93.6 (diketonate CH, C$_o$), 55.6 (CH$_3$, C$_w$), 28.0 (CH$_3$, C$_m$) **Analysis calculated** C 51.11, H 2.75, N 6.11 % **Analysis found** C 50.99, H 2.67, N 5.98 % **ES MS (+)** (CH$_3$OH) m/z 917.10 [MH$^+$] **IR** (cm^{-1}) ν 3082 (w, ArCH), 2964 (w, MeCH), 1743 (w), 1637 (m, CO), 1601 (m, CO), 1494 (m), 1371 (m), 1278 (m), 1200 (s), 1145 (s), 970 (m), 863 (m), 807 (m), 714 (m)

8.13 Synthesis of cobalt(III) *bis*-picolinamide ferrocenyl β-diketonate complexes

The required picolinamide ligand (2 equivalents) and the required ferrocenyl β-diketonate ligand (1 equivalent) in ethanol (10 mL) was added to a solution of cobalt(II) chloride hydrate (1 equivalent) in distilled water (5 mL). Hydrogen peroxide (1 equivalent) was added dropwise and the mixture was heated at 85 °C for 2 hours to give a dark red solution. The solvent was removed under reduced pressure and

the product purified by column chromatography (ethyl acetate/hexane) to give pure product.

8.13.1 Synthesis of CoC$_{42}$H$_{25}$N$_4$O$_4$F$_{14}$Fe (4.10)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a purple solid (0.146 g, 0.142 mmol, 34 %).
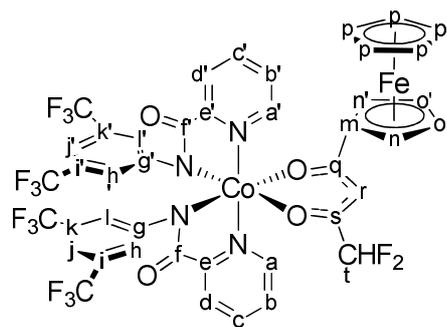
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.1 K) δ 8.22 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 8.18 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.96 (m, 3H, H$_c$, H$_{c'}$ and H$_a$ or H$_{a'}$), 7.76 (d, 1H, H$_a$ or H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.3 Hz), 7.52 (m, 6H, H$_h$, H$_{h'}$, H$_j$, H$_{j'}$, H$_l$ and H$_{l'}$), 7.28 (overlapping ddd, 1H, H$_b$ or H$_{b'}$), 7.18 (overlapping ddd, 1H, H$_b$ or H$_{b'}$), 5.93 (s, 1H, H$_r$), 4.94 (s, 1H, H$_n$ or H$_{n'}$), 4.72 (s, 2H, H$_n$ or H$_{n'}$ and

H$_o$ or H$_{o'}$), 4.62 (s, 1H, H$_o$ or H$_{o'}$), 4.23 (s, 1H, H$_t$), 3.77 (s, 5H, H$_p$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.1 K) δ 193.7 (quaternary diketonate C, C$_s$), 174.0 (quaternary diketonate C, C$_q$), 169.3 (quaternary C-O, C$_f$ or C$_{f'}$), 169.2 (quaternary C-O, C$_f$ or C$_{f'}$), 156.6 (quaternary aniline C, C$_e$ or C$_{e'}$), 156.5 (quaternary aniline C, C$_e$ or C$_{e'}$), 148.8 (quaternary aromatic C, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.5 (quaternary aromatic C, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 147.7 (aniline CH, C$_a$ and C$_{a'}$), 140.5 (aniline CH, C$_c$ or C$_{c'}$), 140.3 (aniline CH, C$_c$ or C$_{c'}$), 132.5 (CHF$_2$, C$_t$), 132.1 (aromatic CH, C$_h$ and C$_l$ or C$_{h'}$ and C$_{l'}$), 131.8 (aromatic CH, C$_h$ and C$_l$ or C$_{h'}$ and C$_{l'}$), 128.5 (CF$_3$), 128.2 (CF$_3$), 127.0 (aniline CH, C$_b$ or C$_{b'}$), 126.7 (aniline CH, C$_b$ or C$_{b'}$), 126.2 (aniline CH, C$_d$ or C$_{d'}$), 126.0 (aniline CH, C$_d$ or C$_{d'}$), 123.9 (quaternary aromatic C, C$_g$ or C$_{g'}$), 121.7 (quaternary aromatic C, C$_g$ or C$_{g'}$), 118.7 (aromatic CH, C$_j$ and C$_{j'}$), 90.9 (diketonate CH, C$_r$), 78.3 (quaternary Cp C, C$_m$), 74.0 (Cp CH, C$_o$ or C$_{o'}$), 73.7 (Cp CH, C$_o$ or C$_{o'}$), 70.4 (Cp CH, C$_p$), 69.7 (Cp CH, C$_n$ or C$_{n'}$), 68.9 (Cp CH, C$_n$ or C$_{n'}$) **Analysis calculated** C 48.96, H 2.45, N 5.44 % **Analysis found** C 49.03, H 2.50, N 5.30 % **ES MS (+)** (CH$_3$OH) *m/z* 1031.05 [MH$^+$] **IR** (cm^{-1}) ν 3080 (w, ArCH), 1636 (m, CO), 1601 (m, CO), 1557 (m, CO), 1376 (s), 1276 (s), 1111 (s), 966 (w), 879 (w), 687 (m), 608 (w), 588 (w), 509 (m)

8.13.2 Synthesis of CoC₄₂H₂₄N₄O₄F₁₅Fe (4.11)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a purple solid (0.200 g, 0.191 mmol, 45 %). Recrystallisation by vapour diffusion (chloroform/pentane) at room temperature yielded orange crystals suitable for single crystal X-ray diffraction.
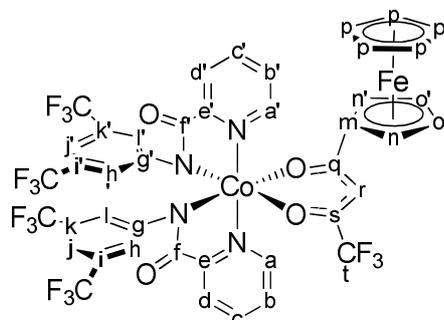
¹H NMR (CDCl₃, 400.13 MHz, 300.0 K) δ 8.34 (d, 1H, H_d or $H_{d'}$, ^3J (^{1}H-^{1}H) = 7.0 Hz), 8.25 (d, 1H, H_d or $H_{d'}$, ^3J (^{1}H-^{1}H) = 6.6 Hz), 8.00 (overlapping td, 2H, H_c and $H_{c'}$), 7.88 (d, 1H, H_a or $H_{a'}$, ^3J (^{1}H-^{1}H) = 4.4 Hz), 7.75 (d, 1H, H_a or $H_{a'}$, ^3J (^{1}H-^{1}H) = 4.0 Hz), 7.54 (s, 2H, H_j and $H_{j'}$), 7.48 (br. s, 4H, H_h, $H_{h'}$, H_l and $H_{l'}$), 7.29 (overlapping ddd, 1H, H_b or $H_{b'}$), 7.20 (overlapping ddd, 1H, H_b or $H_{b'}$), 5.94 (s, 1H, H_r), 4.93 (s, 1H, H_n or $H_{n'}$), 4.76 (s, 1H, H_n or $H_{n'}$), 4.72 (s, 1H, H_o or $H_{o'}$), 4.69 (s, 1H, H_o or $H_{o'}$), 3.79 (s, 5H, H_p) **¹³C {¹H} NMR** (CDCl₃, 100.6 MHz, 300.0 K) δ 195.1 (quaternary diketonate $\underline{C}$, C_s), 185.3 (quaternary diketonate $\underline{C}$, C_q), 167.8 (quaternary $\underline{C}$-O, C_f and $C_{f'}$), 157.0 (quaternary aniline $\underline{C}$, C_e and $C_{e'}$), 148.5 (quaternary aromatic $\underline{C}$, C_i and C_k or $C_{i'}$ and $C_{k'}$), 148.4 (quaternary aromatic $\underline{C}$, C_i and C_k or $C_{i'}$ and $C_{k'}$), 147.6 (aniline $\underline{C}$H, C_a or $C_{a'}$), 147.5 (aniline $\underline{C}$H, C_a or $C_{a'}$), 140.6 (aniline $\underline{C}$H, C_c or $C_{c'}$), 140.5 (aniline $\underline{C}$H, C_c or $C_{c'}$), 132.3 (aromatic $\underline{C}$H, C_h and C_l or $C_{h'}$ and $C_{l'}$), 132.0 (aromatic $\underline{C}$H, C_h and C_l or $C_{h'}$ and $C_{l'}$), 131.8 ($\underline{C}$F₃, C_t), 128.7 ($\underline{C}$F₃), 128.4 ($\underline{C}$F₃), 127.1 (aniline $\underline{C}$H, C_b or $C_{b'}$), 126.8 (aniline $\underline{C}$H, C_b or $C_{b'}$), 126.4 (aniline $\underline{C}$H, C_d or $C_{d'}$), 126.2 (aniline $\underline{C}$H, C_d or $C_{d'}$), 124.1 (quaternary aromatic $\underline{C}$, C_g or $C_{g'}$), 121.4 (quaternary aromatic $\underline{C}$, C_g or $C_{g'}$), 118.7 (aromatic $\underline{C}$H, C_j and $C_{j'}$), 91.2 (diketonate $\underline{C}$H, C_r), 78.2 (quaternary Cp $\underline{C}$, C_m), 74.5 (Cp $\underline{C}$H, C_o or $C_{o'}$), 74.1 (Cp $\underline{C}$H, C_o or $C_{o'}$), 70.6 (Cp $\underline{C}$H, C_p), 69.8 (Cp $\underline{C}$H, C_n or $C_{n'}$), 69.1 (Cp $\underline{C}$H, C_n or $C_{n'}$) **Analysis calculated** (+ 1 H₂O) C 47.30, H 2.46, N 5.25 % **Analysis found** C 47.09, H 2.78, N 5.02 % **ES MS (+)** (CH₃OH) m/z 1049.04 [MH⁺] **IR** (cm⁻¹) ν 3086 (w, ArCH), 2963 (w), 1636 (m, CO), 1572 (m, CO), 1462 (w), 1377 (s), 1277 (m), 1227 (s), 796 (w), 688 (m), 538 (m)

8.13.3 Synthesis of CoC$_{42}$H$_{27}$N$_4$O$_4$F$_{12}$Fe (4.12)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a red solid (0.140 g, 0.141 mmol, 34 %).
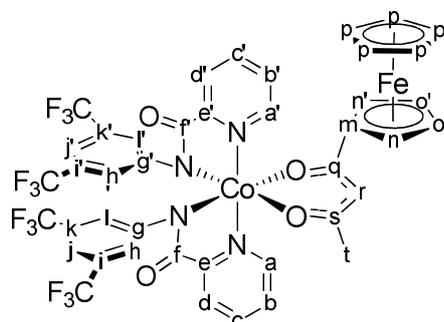
¹H NMR (CDCl$_3$, 500.23 MHz, 300.1 K) δ 8.22 (d, 1H, H$_d$ or H$_{d'}$, ^3J (¹H-¹H) = 7.3 Hz), 8.16 (d, 1H, H$_d$ or H$_{d'}$, ^3J (¹H-¹H) = 7.6 Hz), 7.93 (overlapping td, 2H, H$_c$ and H$_{c'}$, ^3J (¹H-¹H) = 7.4 Hz and ^3J (¹H-¹H) = 1.3 Hz), 7.81 (d, 1H, H$_a$ or H$_{a'}$, ^3J (¹H-¹H) = 5.5 Hz), 7.78 (d, 1H, H$_a$ or H$_{a'}$, ^3J (¹H-¹H) = 5.5 Hz), 7.53 (m, 6H, H$_h$, H$_{h'}$, H$_j$, H$_{j'}$, H$_l$, H$_{l'}$), 7.24 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (¹H-¹H) = 7.5 Hz, ^3J (¹H-¹H) = 5.7 Hz and ^4J (¹H-¹H) = 1.3 Hz), 7.14 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (¹H-¹H) = 7.5 Hz, ^3J (¹H-¹H) = 5.7 Hz and ^4J (¹H-¹H) = 1.2 Hz), 5.61 (s, 1H, H$_r$), 4.88 (s, 1H, H$_n$ or H$_{n'}$), 4.62 (s, 1H, H$_n$ or H$_{n'}$), 4.55 (s, 1H, H$_o$ or H$_{o'}$), 4.47 (s, 1H, H$_o$ or H$_{o'}$), 3.73 (s, 5H, H$_p$), 2.10 (s, 3H, H$_t$) **¹³C {¹H} NMR** (CDCl$_3$, 125.8 MHz, 300.1 K) δ 188.0 (quaternary diketonate C̲, C$_s$), 187.4 (quaternary diketonate C̲, C$_q$), 169.4 (quaternary C̲-O, C$_f$ or C$_{f'}$), 169.2 (quaternary C̲-O, C$_f$ or C$_{f'}$), 157.0 (quaternary aniline C̲, C$_e$ or C$_{e'}$), 156.8 (quaternary aniline C̲, C$_e$ or C$_{e'}$), 148.6 (quaternary aromatic C̲, C$_i$, C$_{i'}$, C$_k$ and C$_{k'}$), 148.2 (aniline C̲H, C$_a$ or C$_{a'}$), 148.0 (aniline C̲H, C$_a$ or C$_{a'}$), 140.1 (aniline C̲H, C$_c$ or C$_{c'}$), 139.9 (aniline C̲H, C$_c$ or C$_{c'}$), 132.0 (aromatic C̲H, C$_h$ and C$_l$ or C$_{h'}$ and C$_{l'}$), 131.7 (aromatic C̲H, C$_h$ and C$_l$ or C$_{h'}$ and C$_{l'}$), 128.2 (C̲F$_3$), 128.0 (C̲F$_3$), 126.5 (aniline C̲H, C$_b$ or C$_{b'}$), 126.3 (aniline C̲H, C$_b$ or C$_{b'}$), 125.9 (aniline C̲H, C$_d$ and C$_{d'}$), 124.0 (quaternary aromatic C̲, C$_g$ or C$_{g'}$), 121.8 (quaternary aromatic C̲, C$_g$ or C$_{g'}$), 118.3 (aromatic C̲H, C$_j$ and C$_{j'}$), 94.1 (diketonate C̲H, C$_r$), 78.9 (quaternary Cp C̲, C$_m$), 72.5 (Cp C̲H, C$_o$ or C$_{o'}$), 72.1 (Cp C̲H, C$_o$ or C$_{o'}$), 70.0 (Cp C̲H, C$_p$), 69.5 (Cp C̲H, C$_n$ or C$_{n'}$), 67.9 (Cp C̲H, C$_n$ or C$_{n'}$), 27.5 (C̲H$_3$, C$_t$) **Analysis calculated** C 50.73, H 2.74, N 5.63 % **Analysis found** C 50.64, H 2.63, N 5.71 % **ES MS (+)** (CH$_3$OH) *m/z* 995.06 [MH$^+$] **IR** (cm^{-1}) ν 3091 (w, ArCH), 1637 (m, CO), 1602 (m, CO), 1509 (m), 1368 (s), 1300 (s), 1225 (s), 1140 (s), 964 (w), 878 (w), 828 (w), 748 (w), 688 (m), 513 (m)

8.13.4 Synthesis of CoC$_{47}$H$_{29}$N$_4$O$_4$F$_{12}$Fe (4.13)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a purple solid (0.151 g, 0.143 mmol, 34 %). Recrystallisation by slow evaporation from methanol at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
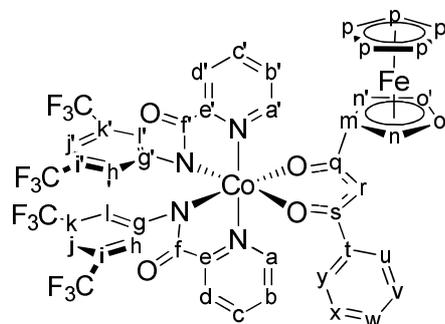
^{1}H NMR (CDCl$_3$, 300.13 MHz, 300.0 K) δ 8.27 (dd, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 0.9 Hz), 8.16 (dd, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 0.8 Hz), 7.95 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz and ^3J (^{1}H-^{1}H) = 1.3 Hz), 7.85 (m, 5H, H$_c$ or H$_{c'}$, H$_a$, H$_{a'}$, H$_j$ and H$_{j'}$), 7.57 (m, 7H, H$_h$, H$_{h'}$, H$_l$, H$_{l'}$, H$_u$, H$_w$ and H$_y$), 7.47 (t, 2H, H$_v$ and H$_x$, ^3J (^{1}H-^{1}H) = 7.0 Hz), 7.16 (overlapping ddd, 2H, H$_b$ and H$_{b'}$, ^4J (^{1}H-^{1}H) = 1.5 Hz), 6.29 (s, 1H, H$_r$), 4.98 (m, 1H, H$_n$ or H$_{n'}$), 4.75 (m, 1H, H$_n$ or H$_{n'}$), 4.62 (td, 1H, H$_o$ or H$_{o'}$, ^3J (^{1}H-^{1}H) = 2.6 Hz and ^3J (^{1}H-^{1}H) = 1.2 Hz), 4.54 (td, 1H, H$_o$ or H$_{o'}$, ^3J (^{1}H-^{1}H) = 2.5 Hz and ^3J (^{1}H-^{1}H) = 1.3 Hz), 3.76 (s, 5H, H$_p$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 189.8 (quaternary diketonate $\underline{C}$, C$_s$), 179.6 (quaternary diketonate $\underline{C}$, C$_q$), 169.2 (quaternary $\underline{C}$-O, C$_f$ and C$_{f'}$), 156.8 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 156.5 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 149.5 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 149.4 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 149.1 (aniline $\underline{C}$H, C$_a$ and C$_{a'}$), 141.0 (aniline $\underline{C}$H, C$_c$ or C$_{c'}$), 140.7 (aniline $\underline{C}$H, C$_c$ or C$_{c'}$), 137.2 (quaternary aromatic $\underline{C}$, C$_t$), 131.9 (aromatic $\underline{C}$H, C$_v$ and C$_x$), 131.3 (aromatic $\underline{C}$H, C$_u$ or C$_y$), 131.0 (aromatic $\underline{C}$H, C$_u$ or C$_y$), 128.6 (aromatic $\underline{C}$H, C$_w$), 128.5 ($\underline{C}$F$_3$), 127.7 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 127.3 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 127.0 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$, C$_l$ and C$_{l'}$), 125.8 (aniline $\underline{C}$H, C$_d$ or C$_{d'}$), 125.6 (aniline $\underline{C}$H, C$_d$ or C$_{d'}$), 124.6 (quaternary aromatic $\underline{C}$, C$_g$ or C$_{g'}$), 121.9 (quaternary aromatic $\underline{C}$, C$_g$ or C$_{g'}$), 117.5 (aromatic $\underline{C}$H, C$_j$ and C$_{j'}$), 91.4 (diketonate $\underline{C}$H, C$_r$), 80.1 (quaternary Cp $\underline{C}$, C$_m$), 72.5 (Cp $\underline{C}$H, C$_o$ or C$_{o'}$), 72.2 (Cp $\underline{C}$H, C$_o$ or C$_{o'}$), 70.0 (Cp $\underline{C}$H, C$_p$), 69.5 (Cp $\underline{C}$H, C$_n$ or C$_{n'}$), 68.4 (Cp $\underline{C}$H, C$_n$ or C$_{n'}$) **Analysis calculated** C 53.43, H 2.77, N 5.30 % **Analysis found** C 53.38, H 2.84, N 5.33 % **ES MS (+)** (CH$_3$OH) *m/z* 1057.08 [MH$^+$] **IR** (cm^{-1}) ν 3080 (w, ArCH), 2070 (w), 1997 (br. w), 1639 (m, CO), 1601 (m, CO), 1530 (m), 1441 (m), 1335 (s), 1228 (s), 1173 (m), 1035 (m), 805 (m), 760 (m), 686 (s), 608 (w), 556 (w), 511 (s)

8.13.5 Synthesis of CoC₄₇H₂₈N₄O₄F₁₃Fe (4.14)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a purple solid (0.245 g, 0.228 mmol, 54 %). Recrystallisation by vapour diffusion (chloroform/pentane) at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
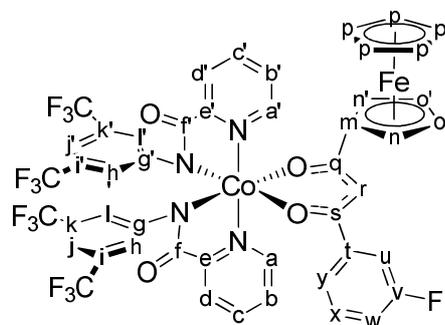
¹H NMR (CDCl₃, 400.13 MHz, 300.0 K) δ 8.27 (d, 1H, H_d or $H_{d'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 8.17 (d, 1H, H_d or $H_{d'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 7.96 (td, 1H, H_c or $H_{c'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.90 (td, 1H, H_c or $H_{c'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.84 (m, 2H, H_a or $H_{a'}$ and H_u), 7.65 (d, 1H, H_a or $H_{a'}$, ^3J (^{1}H-^{1}H) = 7.9 Hz), 7.52 (m, 8H, H_h, $H_{h'}$, H_j, $H_{j'}$, H_l, $H_{l'}$, H_w and H_x), 7.24 (m, 1H, H_y), 7.20 (ddd, 1H, H_b and $H_{b'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.4 Hz), 7.15 (ddd, 1H, H_b and $H_{b'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz, ^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.3 Hz), 6.23 (s, 1H, H_r), 4.98 (m, 1H, H_n or $H_{n'}$), 4.75 (m, 1H, H_n or $H_{n'}$), 4.65 (m, 1H, H_o or $H_{o'}$), 4.56 (m, 1H, H_o or $H_{o'}$), 3.77 (s, 5H, H_p) **¹³C {¹H} NMR** (CDCl₃, 100.6 MHz, 300.0 K) δ 190.3 (quaternary diketonate <u>C</u>, C_s), 178.0 (quaternary diketonate <u>C</u>, C_q), 169.4 (quaternary <u>C</u>-O, C_f or $C_{f'}$), 169.2 (quaternary <u>C</u>-O, C_f or $C_{f'}$), 163.9 (quaternary aromatic <u>C</u>, C_v), 157.0 (quaternary aniline <u>C</u>, C_e or $C_{e'}$), 156.6 (quaternary aniline <u>C</u>, C_e or $C_{e'}$), 148.6 (quaternary aromatic <u>C</u>, C_i, $C_{i'}$, C_k and $C_{k'}$), 148.0 (aniline <u>C</u>H, C_a or $C_{a'}$), 147.9 (aniline <u>C</u>H, C_a or $C_{a'}$), 140.3 (aniline <u>C</u>H, C_c or $C_{c'}$), 140.1 (aniline <u>C</u>H, C_c or $C_{c'}$), 138.9 (quaternary aromatic <u>C</u>, C_t), 132.1 (aromatic <u>C</u>H, C_u or C_w), 132.0 (aromatic <u>C</u>H, C_u or C_w), 131.8 (aromatic <u>C</u>H, C_h, $C_{h'}$, C_l and $C_{l'}$), 130.5 (aromatic <u>C</u>H, C_x), 128.1 (<u>C</u>F₃), 126.8 (aniline <u>C</u>H, C_b or $C_{b'}$), 126.5 (aniline <u>C</u>H, C_b or $C_{b'}$), 126.1 (aniline <u>C</u>H, C_d and $C_{d'}$), 124.2 (quaternary aromatic <u>C</u>, C_g or $C_{g'}$), 122.6 (quaternary aromatic <u>C</u>, C_g or $C_{g'}$), 119.0 (aromatic <u>C</u>H, C_y), 113.7 (aromatic <u>C</u>H, C_j or $C_{j'}$), 113.5 (aromatic <u>C</u>H, C_j or $C_{j'}$), 91.4 (diketonate <u>C</u>H, C_r), 79.4 (quaternary Cp <u>C</u>, C_m), 73.0 (Cp <u>C</u>H, C_o or $C_{o'}$), 72.6 (Cp <u>C</u>H, C_o or $C_{o'}$), 70.1 (Cp <u>C</u>H, C_p), 69.7 (Cp <u>C</u>H, C_n or $C_{n'}$), 68.1 (Cp <u>C</u>H, C_n or $C_{n'}$) **Analysis calculated** C 52.54, H 2.63, N 5.21 % **Analysis found** C 52.47, H 2.56, N 5.20 % **ES MS (+)** (CH₃OH) *m/z* 1075.07 [MH⁺] **IR** (cm⁻¹) ν 3084 (w, ArCH), 1633 (m, CO), 1601 (m, CO), 1529 (m), 1388 (m), 1309 (s), 1173 (m), 1120(s), 956 (w), 762 (w), 703 (m), 608 (m)

8.13.6 Synthesis of CoC$_{47}$H$_{28}$N$_4$O$_4$F$_{13}$Fe (4.15)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a purple solid (0.273 g, 0.254 mmol, 61 %). Recrystallisation by vapour diffusion (chloroform/pentane) at room temperature yielded orange crystals suitable for single crystal X-ray diffraction.
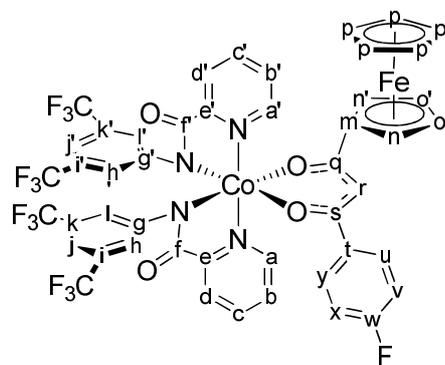
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.1 K) δ 8.27 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 8.17 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz), 7.96 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.4 Hz and ^3J (^{1}H-^{1}H) = 1.2 Hz), 7.90 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.4 Hz and ^3J (^{1}H-^{1}H) = 1.3 Hz), 7.85 (m, 4H, H$_a$, H$_{a'}$, H$_v$ and H$_x$), 7.58 (br. s, 4H, H$_h$, H$_{h'}$, H$_l$ and H$_{l'}$), 7.52 (s, 2H, H$_j$ and H$_{j'}$), 7.19 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz,

^3J (^{1}H-^{1}H) = 5.7 Hz and ^4J (^{1}H-^{1}H) = 1.4 Hz), 7.14 (m, 3H, H$_b$ or H$_{b'}$, H$_u$ and H$_y$), 6.22 (s, 1H, H$_r$), 4.97 (s, 1H, H$_n$ or H$_{n'}$), 4.74 (s, 1H, H$_n$ or H$_{n'}$), 4.63 (s, 1H, H$_o$ or H$_{o'}$), 4.54 (s, 1H, H$_o$ or H$_{o'}$), 3.76 (s, 5H, H$_p$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 299.9 K) δ 188.6 (quaternary diketonate C̲, C$_s$), 178.4 (quaternary diketonate C̲, C$_q$), 169.4 (quaternary C̲-O, C$_f$ and C$_{f'}$), 161.0 (quaternary aromatic C̲, C$_w$), 157.0 (quaternary aniline C̲, C$_e$ and C$_{e'}$), 148.6 (quaternary aromatic C̲, C$_i$, C$_{i'}$, C$_k$ and C$_{k'}$), 148.1 (aniline C̲H, C$_a$ and C$_{a'}$), 140.3 (aniline C̲H, C$_c$ or C$_{c'}$), 140.1 (aniline C̲H, C$_c$ or C$_{c'}$), 132.6 (quaternary aromatic C̲, C$_t$), 129.2 (aromatic C̲H, C$_u$ and C$_y$), 129.0 (aromatic C̲H, C$_v$ and C$_x$), 128.2 (C̲F$_3$), 126.7 (aromatic C̲H, C$_h$, C$_{h'}$, C$_l$ and C$_{l'}$), 126.4 (aniline C̲H, C$_b$ and C$_{b'}$), 126.1 (aniline C̲H, C$_d$ and C$_{d'}$), 124.0 (quaternary aromatic C̲, C$_g$ or C$_{g'}$), 121.8 (quaternary aromatic C̲, C$_g$ or C$_{g'}$), 116.0 (aromatic C̲H, C$_j$ or C$_{j'}$), 115.8 (aromatic C̲H, C$_j$ or C$_{j'}$), 90.9 (diketonate C̲H, C$_r$), 79.6 (quaternary Cp C̲, C$_m$), 72.8 (Cp C̲H, C$_o$ or C$_{o'}$), 72.4 (Cp C̲H, C$_o$ or C$_{o'}$), 70.1 (Cp C̲H, C$_p$), 69.6 (Cp C̲H, C$_n$ or C$_{n'}$), 68.0 (Cp C̲H, C$_n$ or C$_{n'}$) **Analysis calculated** C 52.54, H 2.63, N 5.21 % **Analysis found** C 52.51, H 2.66, N 5.27 % **ES MS (+)** (CH$_3$OH) *m/z* 1075.07 [MH$^+$] **IR** (cm^{-1}) ν 3085 (w, ArCH), 1636 (m, CO), 1599 (m, CO), 1521 (m), 1497 (m), 1369 (m), 1274 (s), 1119 (s), 1101 (s), 1003 (m), 895 (m), 686 (m), 608 (w) 498 (s)

8.13.7 Synthesis of CoC$_{47}$H$_{27}$N$_4$O$_4$F$_{14}$Fe (4.16)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a purple solid (0.263 g, 0.240 mmol, 57 %). Recrystallisation by slow evaporation from methanol at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
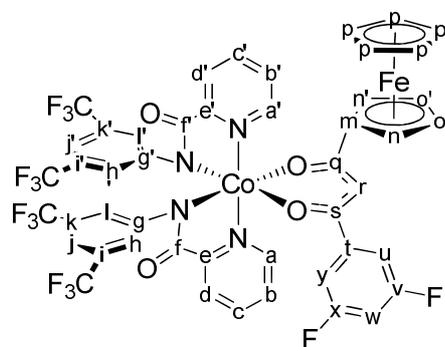
^{1}H NMR (CDCl$_3$, 500.23 MHz, 300.0 K) δ 8.28 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz), 8.18 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.4 Hz), 7.98 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.91 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.83 (overlapping d, 2H, H$_a$ and H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 7.57 (br. s, 4H, H$_h$, H$_{h'}$ and H$_l$ and H$_{l'}$), 7.53 (s, 2H, H$_j$ and H$_{j'}$), 7.32 (m, 2H, H$_u$

and H$_y$), 7.23 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.4 Hz, ^3J (^{1}H-^{1}H) = 5.9 Hz and ^4J (^{1}H-^{1}H) = 1.4 Hz), 7.16 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.4 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.4 Hz), 6.99 (m, 1H, H$_w$), 6.17 (s, 1H, H$_r$), 4.99 (s, 1H, H$_n$ or H$_{n'}$), 4.76 (s, 1H, H$_n$ or H$_{n'}$), 4.67 (s, 1H, H$_o$ or H$_{o'}$), 4.59 (s, 1H, H$_o$ or H$_{o'}$), 3.77 (s, 5H, H$_p$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 299.9 K) δ 191.2 (quaternary diketonate $\underline{C}$, C$_s$), 176.5 (quaternary diketonate $\underline{C}$, C$_q$), 169.3 (quaternary $\underline{C}$-O, C$_f$ or C$_{f'}$), 169.2 (quaternary $\underline{C}$-O, C$_f$ or C$_{f'}$), 164.5 (quaternary aromatic $\underline{C}$, C$_v$ and C$_x$), 156.9 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 156.6 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 148.5 (quaternary aromatic $\underline{C}$, C$_i$, C$_{i'}$, C$_k$ and C$_{k'}$), 147.9 (aniline $\underline{C}$H, C$_a$ or C$_{a'}$), 147.8 (aniline $\underline{C}$H, C$_a$ or C$_{a'}$), 140.4 (aniline $\underline{C}$H, C$_c$ or C$_{c'}$), 140.2 (aniline $\underline{C}$H, C$_c$ or C$_{c'}$), 134.2 (quaternary aromatic $\underline{C}$, C$_t$), 128.2 ($\underline{C}$F$_3$), 126.8 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$, C$_l$ and C$_{l'}$), 126.6 (aniline $\underline{C}$H, C$_b$ and C$_{b'}$), 126.2 (aniline $\underline{C}$H, C$_d$ and C$_{d'}$), 123.9 (quaternary aromatic $\underline{C}$, C$_g$ or C$_{g'}$), 121.7 (quaternary aromatic $\underline{C}$, C$_g$ or C$_{g'}$), 116.0 (aromatic $\underline{C}$H, C$_j$ or C$_{j'}$), 120.8 (aromatic $\underline{C}$H, C$_j$ or C$_{j'}$), 118.5 (aromatic $\underline{C}$H, C$_j$ or C$_{j'}$), 109.8 (aromatic $\underline{C}$H, C$_u$ and C$_y$), 109.6 (aromatic $\underline{C}$H, C$_u$ and C$_y$), 107.2 (aromatic $\underline{C}$H, C$_w$), 91.6 (diketonate $\underline{C}$H, C$_r$), 79.2 (quaternary Cp $\underline{C}$, C$_m$), 73.3 (Cp $\underline{C}$H, C$_o$ or C$_{o'}$), 72.9 (Cp $\underline{C}$H, C$_o$ or C$_{o'}$), 70.2 (Cp $\underline{C}$H, C$_p$), 69.7 (Cp $\underline{C}$H, C$_n$ or C$_{n'}$), 68.2 (Cp $\underline{C}$H, C$_n$ or C$_{n'}$) **Analysis calculated** C 51.67, H 2.49, N 5.13 % **Analysis found** C 51.52, H 2.56, N 5.18 % **ES MS (+)** (CH$_3$OH) *m/z* 1093.06 [MH$^+$] **IR** (cm^{-1}) ν 3056 (w, ArCH), 1633 (m, CO), 1602 (m, CO), 1529 (m), 1367 (m), 1276 (s), 1116 (s), 964 (m), 879 (w), 762 (w), 703 (m), 608 (w), 502 (m)

8.13.8 Synthesis of CoC$_{48}$H$_{31}$N$_4$O$_4$F$_{12}$Fe (4.17)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a red solid (0.273 g, 0.254 mmol, 61 %). Recrystallisation by slow evaporation from ethyl acetate at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
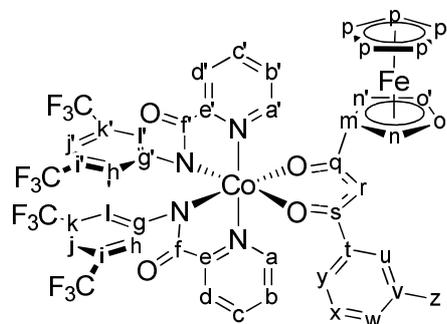
^{1}H NMR (CDCl$_3$, 400.13 MHz, 300.0 K) δ 8.26 (dd, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.8 Hz and ^3J (^{1}H-^{1}H) = 1.0 Hz), 8.15 (dd, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.8 Hz and ^3J (^{1}H-^{1}H) = 1.0 Hz), 7.95 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.3 Hz), 7.87 (m, 3H, H$_c$ or H$_{c'}$, H$_a$ and H$_{a'}$), 7.69 (s, 1H, H$_u$), 7.64 (d, 1H, H$_y$, ^3J (^{1}H-^{1}H) = 7.3 Hz), 7.58 (br. s, 4H, H$_h$, H$_{h'}$, H$_l$ and H$_{l'}$), 7.51 (s, 2H, H$_j$ and H$_{j'}$), 7.36 (m, 2H,

H$_w$ and H$_x$), 7.19 (ddd, 1H, H$_b$ and H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.4 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.5 Hz), 7.13 (ddd, 1H, H$_b$ and H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.4 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.5 Hz), 6.29 (s, 1H, H$_r$), 4.99 (m, 1H, H$_n$ or H$_{n'}$), 4.76 (m, 1H, H$_n$ or H$_{n'}$), 4.62 (td, 1H, H$_o$ or H$_{o'}$, ^3J (^{1}H-^{1}H) = 2.5 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 4.53 (td, 1H, H$_o$ or H$_{o'}$, ^3J (^{1}H-^{1}H) = 2.5 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 3.75 (s, 5H, H$_p$), 2.46 (s, 3H, H$_z$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 100.6 MHz, 300.0 K) δ 189.2 (quaternary diketonate C̲, C$_s$), 180.0 (quaternary diketonate C̲, C$_q$), 169.4 (quaternary C̲-O, C$_f$ or C$_{f'}$), 169.3 (quaternary C̲-O, C$_f$ or C$_{f'}$), 157.0 (quaternary aniline C̲, C$_e$ or C$_{e'}$), 156.7 (quaternary aniline C̲, C$_e$ or C$_{e'}$), 148.7 (quaternary aromatic C̲, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.6 (quaternary aromatic C̲, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.1 (aniline C̲H, C$_a$ or C$_{a'}$), 148.0 (aniline C̲H, C$_a$ or C$_{a'}$), 140.2 (aniline C̲H, C$_c$ or C$_{c'}$), 140.0 (aniline C̲H, C$_c$ or C$_{c'}$), 138.9 (quaternary aromatic C̲, C$_v$), 136.7 (quaternary aromatic C̲, C$_t$), 133.0 (aromatic C̲H, C$_w$), 132.0 (aromatic C̲H, C$_u$ or C$_y$), 131.7 (aromatic C̲H, C$_u$ or C$_y$), 128.6 (aromatic C̲H, C$_x$), 128.1 (C̲F$_3$), 127.4 (aromatic C̲H, C$_h$, C$_{h'}$, C$_l$ and C$_{l'}$), 126.7 (aniline C̲H, C$_b$ or C$_{b'}$), 126.4 (aniline C̲H, C$_b$ or C$_{b'}$), 126.0 (aniline C̲H, C$_d$ and C$_{d'}$), 124.2 (quaternary aromatic C̲, C$_g$ or C$_{g'}$), 121.6 (quaternary aromatic C̲, C$_g$ or C$_{g'}$), 118.3 (aromatic C̲H, C$_j$ and C$_{j'}$), 91.2 (diketonate C̲H, C$_r$), 79.7 (quaternary Cp C̲, C$_m$), 72.7 (Cp C̲H, C$_o$ or C$_{o'}$), 72.3 (Cp C̲H, C$_o$ or C$_{o'}$), 70.1 (Cp C̲H, C$_p$), 69.6 (Cp C̲H, C$_n$ or C$_{n'}$), 68.0 (Cp C̲H, C$_n$ or C$_{n'}$), 21.4 (C̲H$_3$, C$_z$) **Analysis calculated** C 53.85, H 2.92, N 5.23 % **Analysis found** C 53.69, H 2.98, N 5.19 % **ES MS (+)** (CH$_3$OH) *m/z* 1071.09 [MH$^+$] **IR** (cm^{-1}) ν 3053 (w, ArCH), 2923 (w, MeCH), 1722 (w), 1634 (m, CO), 1599 (m, CO), 1511 (m), 1365 (m), 1275 (s), 1170 (m), 1121 (s), 1002 (w), 843 (w), 766 (m), 688 (m), 608 (w), 514 (m)

8.13.9 Synthesis of CoC₄₈H₃₁N₄O₄F₁₂Fe (4.18)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a purple solid (0.210 g, 0.197 mmol, 47 %). Recrystallisation by slow evaporation from methanol at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
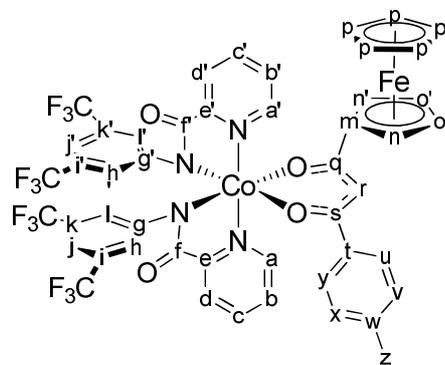
¹H NMR (CDCl₃, 400.13 MHz, 300.0 K) δ 8.26 (dd, 1H, H_d or $H_{d'}$, ^3J (^1H-^1H) = 7.7 Hz and ^3J (^1H-^1H) = 0.9 Hz), 8.15 (dd, 1H, H_d or $H_{d'}$, ^3J (^1H-^1H) = 7.7 Hz and ^3J (^1H-^1H) = 0.9 Hz), 7.94 (td, 1H, H_c or $H_{c'}$, ^3J (^1H-^1H) = 7.7 Hz and ^3J (^1H-^1H) = 1.3 Hz), 7.87 (m, 3H, H_c or $H_{c'}$, H_a and $H_{a'}$), 7.78 (d, 2H, H_u and H_y, ^3J (^1H-^1H) = 8.3 Hz), 7.59 (br. s, 4H, H_h, $H_{h'}$, H_l and $H_{l'}$), 7.51 (s, 2H, H_j and $H_{j'}$), 7.27 (d, 2H, H_v and H_x, ^3J (^1H-^1H) = 8.1 Hz), 7.17 (ddd, 1H, H_b and $H_{b'}$, ^3J (^1H-^1H) = 7.4 Hz, ^3J (^1H-^1H) = 5.8 Hz and ^4J (^1H-^1H) = 1.5 Hz), 7.13 (ddd, 1H, H_b and $H_{b'}$, ^3J (^1H-^1H) = 7.4 Hz, ^3J (^1H-^1H) = 5.8 Hz and ^4J (^1H-^1H) = 1.5 Hz), 6.28 (s, 1H, H_r), 4.97 (m, 1H, H_n or $H_{n'}$), 4.74 (m, 1H, H_n or $H_{n'}$), 4.60 (td, 1H, H_o or $H_{o'}$, ^3J (^1H-^1H) = 2.5 Hz and ^3J (^1H-^1H) = 1.3 Hz), 4.52 (td, 1H, H_o or $H_{o'}$, ^3J (^1H-^1H) = 2.5 Hz and ^3J (^1H-^1H) = 1.3 Hz), 3.75 (s, 5H, H_p), 2.24 (s, 3H, H_z) **¹³C {¹H} NMR** (CDCl₃, 100.6 MHz, 300.0 K) δ 189.9 (quaternary diketonate $\underline{C}$, C_s), 179.7 (quaternary diketonate $\underline{C}$, C_q), 169.4 (quaternary $\underline{C}$-O, C_f or $C_{f'}$), 169.3 (quaternary $\underline{C}$-O, C_f or $C_{f'}$), 157.0 (quaternary aniline $\underline{C}$, C_e or $C_{e'}$), 156.7 (quaternary aniline $\underline{C}$, C_e or $C_{e'}$), 148.7 (quaternary aromatic $\underline{C}$, C_i and C_k or $C_{i'}$ and $C_{k'}$), 148.6 (quaternary aromatic $\underline{C}$, C_i and C_k or $C_{i'}$ and $C_{k'}$), 148.1 (aniline $\underline{C}H$, C_a or $C_{a'}$), 148.0 (aniline $\underline{C}H$, C_a or $C_{a'}$), 143.2 (quaternary aromatic $\underline{C}$, C_w), 140.2 (aniline $\underline{C}H$, C_c or $C_{c'}$), 140.0 (aniline $\underline{C}H$, C_c or $C_{c'}$), 134.0 (quaternary aromatic $\underline{C}$, C_t), 132.0 (aromatic $\underline{C}H$, C_u or C_y), 131.7 (aromatic $\underline{C}H$, C_u or C_y), 129.5 (aromatic $\underline{C}H$, C_v and C_x), 128.1 ($\underline{C}F_3$), 126.9 (aromatic $\underline{C}H$, C_h, $C_{h'}$, C_l and $C_{l'}$), 126.6 (aniline $\underline{C}H$, C_b or $C_{b'}$), 126.4 (aniline $\underline{C}H$, C_b or $C_{b'}$), 126.0 (aniline $\underline{C}H$, C_d and $C_{d'}$), 124.2 (quaternary aromatic $\underline{C}$, C_g or $C_{g'}$), 121.6 (quaternary aromatic $\underline{C}$, C_g or $C_{g'}$), 118.3 (aromatic $\underline{C}H$, C_j and $C_{j'}$), 90.8 (diketonate $\underline{C}H$, C_r), 79.7 (quaternary Cp $\underline{C}$, C_m), 72.6 (Cp $\underline{C}H$, C_o or $C_{o'}$), 72.2 (Cp $\underline{C}H$, C_o or $C_{o'}$), 70.0 (Cp $\underline{C}H$, C_p), 69.6 (Cp $\underline{C}H$, C_n or $C_{n'}$), 67.9 (Cp $\underline{C}H$, C_n or $C_{n'}$), 21.5 ($\underline{C}H_3$, C_z) **Analysis calculated** C 53.85, H 2.92, N 5.23 % **Analysis found** C 53.95, H 2.85, N 5.17 % **ES MS (+)** (CH₃OH) *m/z* 1071.09 [MH⁺] **IR** (cm⁻¹) ν 3081 (w, ArCH), 2924 (w, MeCH), 2853 (w), 1635 (M, CO), 1601 (m, CO), 1495 (m), 1365 (m), 1278 (s), 1229 (w), 1169 (s), 1116 (s), 947 (m), 688 (m), 512 (m)

8.13.10 Synthesis of CoC$_{49}$H$_{33}$N$_4$O$_4$F$_{12}$Fe (4.19)

Purified by column chromatography (ethyl acetate/hexane 1:1) to give a purple solid (0.299 g, 0.276 mmol, 66 %). Recrystallisation by slow evaporation from methanol at room temperature yielded red crystals suitable for single crystal X-ray diffraction.
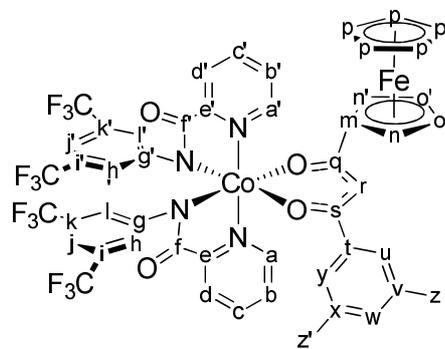
^{1}H NMR (CDCl$_3$, 500.23 MHz, 299.9 K) δ 8.26 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.1 Hz), 8.14 (d, 1H, H$_d$ or H$_{d'}$, ^3J (^{1}H-^{1}H) = 7.1 Hz), 7.94 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.7 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.91 (d, 1H, H$_a$ or H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.5 Hz), 7.87 (td, 1H, H$_c$ or H$_{c'}$, ^3J (^{1}H-^{1}H) = 7.6 Hz and ^3J (^{1}H-^{1}H) = 1.1 Hz), 7.85 (d, 1H, H$_a$ or H$_{a'}$, ^3J (^{1}H-^{1}H) = 5.7 Hz), 7.58 (br. s, 4H, H$_h$, H$_{h'}$ and H$_l$ and H$_{l'}$), 7.51 (s, 2H,

H$_j$ and H$_{j'}$), 7.47 (s, 2H, H$_u$ and H$_y$), 7.21 (s, 1H, H$_w$), 7.19 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.4 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.1 Hz), 7.13 (ddd, 1H, H$_b$ or H$_{b'}$, ^3J (^{1}H-^{1}H) = 7.5 Hz, ^3J (^{1}H-^{1}H) = 5.8 Hz and ^4J (^{1}H-^{1}H) = 1.1 Hz), 6.29 (s, 1H, H$_r$), 4.99 (s, 1H, H$_n$ or H$_{n'}$), 4.77 (s, 1H, H$_n$ or H$_{n'}$), 4.61 (s, 1H, H$_o$ or H$_{o'}$), 4.52 (s, 1H, H$_o$ or H$_{o'}$), 3.74 (s, 5H, H$_p$), 2.41 (s, 6H, H$_z$ and H$_{z'}$) **^{13}C {^{1}H} NMR** (CDCl$_3$, 125.8 MHz, 300.0 K) δ 189.0 (quaternary diketonate $\underline{C}$, C$_s$), 180.2 (quaternary diketonate $\underline{C}$, C$_q$), 169.3 (quaternary $\underline{C}$-O, C$_f$ and C$_{f'}$), 157.0 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 156.7 (quaternary aniline $\underline{C}$, C$_e$ or C$_{e'}$), 148.8 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.6 (quaternary aromatic $\underline{C}$, C$_i$ and C$_k$ or C$_{i'}$ and C$_{k'}$), 148.0 (aniline $\underline{C}$H, C$_a$ and C$_{a'}$), 140.2 (aniline $\underline{C}$H, C$_c$ or C$_{c'}$), 139.9 (aniline $\underline{C}$H, C$_c$ or C$_{c'}$), 138.6 (quaternary aromatic $\underline{C}$, C$_v$ and C$_x$), 136.8 (quaternary aromatic $\underline{C}$, C$_t$), 134.0 (aromatic $\underline{C}$H, C$_u$ and C$_y$), 131.7 (aromatic $\underline{C}$H, C$_w$), 128.1 ($\underline{C}$F$_3$), 126.6 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 126.4 (aniline $\underline{C}$H, C$_b$ or C$_{b'}$), 126.0 (aniline $\underline{C}$H, C$_d$ and C$_{d'}$), 124.6 (aromatic $\underline{C}$H, C$_h$, C$_{h'}$, C$_l$ and C$_{l'}$), 124.0 (quaternary aromatic $\underline{C}$, C$_g$ or C$_{g'}$), 121.8 (quaternary aromatic $\underline{C}$, C$_g$ or C$_{g'}$), 118.3 (aromatic $\underline{C}$H, C$_j$ and C$_{j'}$), 91.2 (diketonate $\underline{C}$H, C$_r$), 79.7 (quaternary Cp $\underline{C}$, C$_m$), 72.6 (Cp $\underline{C}$H, C$_o$ or C$_{o'}$), 72.2 (Cp $\underline{C}$H, C$_o$ or C$_{o'}$), 70.0 (Cp $\underline{C}$H, C$_p$), 69.6 (Cp $\underline{C}$H, C$_n$ or C$_{n'}$), 68.0 (Cp $\underline{C}$H, C$_n$ or C$_{n'}$), 21.2 ($\underline{C}$H$_3$, C$_z$ and C$_{z'}$) **Analysis calculated** (+ 1 H$_2$O) C 53.38, H 3.20, N 5.08 % **Analysis found** C 53.06, H 3.02, N 5.18 % **ES MS (+)** (CH$_3$OH) *m/z* 1085.11 [MH$^+$] **IR** (cm^{-1}) ν 3051 (w, ArCH), 2923 (w, MeCH), 1722 (w), 1633 (m, CO), 1599 (m, CO), 1505 (m), 1462 (w), 1364 (m), 1274 (s), 1176 (m), 1122 (s), 1022 (w) 894 (w), 766 (m), 688 (m), 511 (m)

8.14 Cytotoxic evaluation

Sterile techniques were used throughout this work. Chemicals were purchased from Sigma-Aldrich Chemical Co. MIA PaCa-2 (human pancreatic carcinoma), BE (human colon carcinoma) and ARPE-19 (human retinal pigment epithelial cells) cells were obtained from Prof. Roger Phillips (University of Huddersfield). Stock cell cultures were grown in T-25 or T-75 flasks containing RPMI-1640 complete cell medium (20 mL) and incubated at 37 °C in an atmosphere of 5 % carbon dioxide. The complete medium was prepared from RPMI-1640 incomplete medium (500 mL), sodium pyruvate (5 mL, 0.5 mmol), L-glutamine (5 mL, 1.0 mmol) and foetal bovine serum (FBS) (50 mL). Hank's balanced salt solution (HBSS) was used to wash cells before use and 0.25 % trypsin-EDTA solution was used to detach cells from the flask. MTT stock solutions (5 mg mL^{-1}) were prepared by dissolving MTT (250 µg) in distilled water (50 mL) followed by passage through a 0.2 µm sterile filter. RPMI-1640 incomplete medium, RPMI-1640 complete medium, sodium pyruvate, MTT and MTT stock solutions were all stored at 4 °C. L-glutamine, FBS and 0.25 % trypsin-EDTA solution were all stored at -20 °C. All chemicals except the MTT stock solution were incubated at 37 °C prior to use.

Cells were washed with HBSS (3 × 10 mL) and HBSS carefully removed. 0.25 % trypsin-EDTA solution (5 mL) was added and the flasks incubated at 37 °C for 5 minutes. Following cellular detachment from the flask wall, cell media (10 mL) was added. Cells were split into new flasks, diluted with cell media, the lids loosened and the flasks transferred into the incubator at 37 °C. Each cell suspension (10 µL) was transferred to each side of the glass haemocytometer and cells were counted using an optical microscope and an average taken with units of 10^4 cells mL^{-1}.

8.14.1 The five-day MTT assay (normoxic conditions)

The cell suspension was diluted with RPMI-1640 complete media to give a concentration of 2 × 10^4 cells mL^{-1}. 100 µL of cell media was added to the first lane of the 96-well plate to act as a blank. 100 µL of diluted cell suspension were added to the other wells and the plate incubated at 37 °C overnight with 5 % CO$_2$. Complexes were prepared in DMSO and then further diluted with RPMI-1640 media to give a final concentration of 250 µM (with a final DMSO concentration at 0.1 %). Further dilutions were carried out to give eight different drug concentrations. All experiments were carried out in triplicate. Drug solutions were added to the cells and the plate incubated for five days at 37 °C with 5 % CO$_2$. After five days, 20 µL of a MTT stock solution (5 mg mL^{-1}) was added to each well and incubated for a further

3 hours at 37 °C with 5 % CO_2. The media and MTT was removed and 150 µL of DMSO added to each well to dissolve any formazan crystals. The absorbance at 540 nm was determined with a Thermo Scientific Multiskan EX microplate photometer. Cell survival was determined at the absorbance of treated cells compared to the absorbance of non-treated cells (negative control). IC_{50} values were determined from plots of percentage survival against drug concentration.

8.14.2 The five-day MTT assay (hypoxic conditions)

The assay was conducted according to the protocol stated previously for normoxic conditions. However, all the incubations periods, the addition of the drug and the addition of the MTT solution were carried out inside a Don Whitley Scientific H35 Hypoxystation with the oxygen level set at 0.1 %. Cell culture media was conditioned for 24 hours at 0.1 % O_2 prior to the start of the experiment.

8.14.3 Cell viability assay

Cells were seeded at 1.66×10^5 cells ml^{-1} and incubated at 37 °C overnight with 5 % CO_2. Cells were washed with phosphate-buffered saline (PBS) (3 × 5 mL) and cell media (5 mL) added following cellular detachment. Cell suspensions samples were added to Via-1 cassettes preloaded with acridine orange and 4',6-diamidino-2-phenylindole (DAPI) and analysed with a Chemometec NucleoCounter NC-3000.

8.14.4 Cell cycle assay

Cell suspensions from the cell viability assay were diluted with PBS to 1×10^6 cells ml^{-1} and centrifuged at 400 g min^{-1} for 5 minutes. The supernatant was removed, washed with PBS, centrifuged at 400 g min^{-1} for 5 minutes and PBS removed. Cells were resuspended in lysis buffer (250 µL) supplemented with DAPI (10 µg ml^{-1}) and incubated at 37 °C for 5 minutes. Stabilisation buffer (250 µL) was added and cells analysed with a Chemometec NucleoCounter NC-3000.

8.15 Anti-bacterial evaluation

Anti-bacterial evaluation was performed by The Community for Antimicrobial Drug Discovery (CO-ADD) at The University of Queensland, funded by The Wellcome Trust.

Complexes were prepared in DMSO and water to a give a final concentration of 32 µg mL^{-1} in 384-well non-binding surface (NBS) plate. The final DMSO concentration was at a maximum of 1 % DMSO. All bacteria were cultured in Cation-adjusted Mueller Hinton broth (CAMHB) at 37 °C overnight. A sample of each culture was diluted 40-fold in fresh broth and incubated at 37 °C for 1.5 - 3 hours. The resultant mid-log phase cultures were diluted (CFU mL^{-1} measured by OD$_{600}$), then added to each well of the compound containing plates, giving a cell density of 5×10^5 CFU mL^{-1} and a total volume of 50 µL. Colistin and vancomycin were used as positive bacterial inhibitor standards for Gram-negative and Gram-positive bacteria, respectively. The antibiotics were provided at four concentrations, with two above and two below the MIC value. All the plates were covered and incubated at 37 °C for 18 hours without shaking. All experiments were carried out in duplicate. Inhibition of bacterial growth was determined measuring absorbance at 600 nm (OD$_{600}$), using a Tecan M1000 Pro monochromator plate reader. The percentage of growth inhibition was calculated for each well, using the negative control (media only) and positive control (bacteria without inhibitors) on the same plate as references. The significance of the inhibition values was determined by modified Z-scores, calculated using the median and MAD of the samples (no controls) on the same plate. Samples with inhibition value above 80 % and Z-Score above 2.5 for either replicate were classed as actives. Samples with inhibition values in the range 50 to 80 % and Z-Score above 2.5 for either replicate were classed as partial actives.

8.16 Anti-fungal evaluation

Anti-fungal evaluation was performed by The Community for Antimicrobial Drug Discovery (CO-ADD) at The University of Queensland, funded by The Wellcome Trust.

8.16.1 Anti-fungal screening procedure

Complexes were prepared in DMSO and water to a give a final concentration of 32 µg mL^{-1} in 384-well NBS plate. The final DMSO concentration was at a maximum of 1 % DMSO. Fungal strains were cultured for three days on yeast extract-peptone dextrose (YPD) agar at 30 °C. A yeast suspension of 1×10^6 to 5×10^6 CFU mL^{-1} (as

determined by OD_{530}) was prepared from five colonies. The suspension was diluted and added to each well of the compound-containing plates giving a final cell density of fungi suspension of 2.5×10^3 CFU mL^{-1} and a total volume of 50 µL. Fluconazole was used as a positive fungal inhibitor standard. All the plates were covered and incubated at 35 °C for 24 hours without shaking. All experiments were carried out in duplicate. Growth inhibition of *C. albicans* was determined measuring absorbance at 530 nm (OD_{530}) and the growth inhibition of *C. neoformans* was determined measuring the difference in absorbance between 600 and 570 nm ($OD_{600-570}$), after the addition of 0.001 % resazurin and incubation at 35 °C for an additional 2 hours. The absorbance was measured using a Biotek Synergy HTX plate reader. The percentage of growth inhibition was calculated for each well, using the negative control (media only) and positive control (fungi without inhibitors) on the same plate. The significance of the inhibition values was determined by modified Z-scores, calculated using the median and MAD of the samples (no controls) on the same plate. Samples with inhibition value above 80 % and Z-Score above 2.5 for either replicate were classed as actives. Samples with inhibition values in the range 50 to 80 % and Z-Score above 2.5 for either replicate were classed as partial actives.

8.16.2 Anti-fungal hit confirmation procedure

Complexes were prepared in DMSO and water to a give a final concentration of 32 µg mL^{-1} and serially diluted two fold for eight times. Each sample concentration was prepared in 384-well non-binding surface plate for each fungal strain or tissue-culture treated plates for mammalian cell types. The final DMSO concentration was at a maximum of 0.5 % DMSO. Fungal strains were cultured for three days on YPD agar at 30 °C. A yeast suspension of 1×10^6 to 5×10^6 CFU mL^{-1} (as determined by OD_{530}) was prepared from five colonies. The suspension was subsequently diluted and added to each well of the compound-containing plates giving a final cell density of fungi suspension of 2.5×10^3 CFU mL^{-1} and a total volume of 50 µL. Fluconazole was used as a positive fungal inhibitor standard. The anti-fungal was provided at four concentrations, with two above and two below the MIC value. All plates were covered and incubated at 35 °C for 36 hours without shaking. All experiments were performed in duplicate. Growth inhibition of *C. albicans* was determined measuring absorbance at 630 nm (OD_{630}) and the growth inhibition of *C. neoformans* was determined measuring the difference in absorbance between 600 and 570 nm ($OD_{600-570}$) after the addition of 0.001 % resazurin and incubation at 35 °C for 2 hours. The absorbance was measured using a Biotek Multiflo Synergy HTX plate reader. The percentage of growth inhibition was calculated for each well, using the negative control (media only) and positive control (fungi without inhibitors) on

the same plate. The MIC was determined as the lowest concentration at which the growth was fully inhibited, defined by an inhibition ≥ 80% for *C. albicans* and an inhibition ≥ 70% for *C. neoformans.* Due to a higher variance in growth and inhibition, a lower threshold was applied to the data for *C. neoformans*. In addition, the maximal percentage of growth inhibition is reported as DMax, indicating any compounds with marginal activity. Hits were classified by MIC ≤ 16 μg mL^{-1} or MIC ≤ 10 μM in either replicate.

To assess the cytotoxicity, HEK293 cells were counted manually in a Neubauer haemocytometer and then plated in the 384-well plates containing the compounds to give a density of 6000 cells well^{-1} in a final volume of 50 μL. Dulbecco's modified eagle medium (DMEM) supplemented with 10 % FBS was used as growth media and the cells were incubated together with the compounds for 20 hours at 37 °C in 5 % CO_2. Tamoxifen was used as a positive cytotoxicity standard. The cytotoxic drug was provided at four concentrations, with two above and two below the CC_{50} value. All experiments were performed in duplicate. Cytotoxicity was measured by fluorescence with excitation at 560 nm and emission at 590 nm ($F_{560/590}$), after addition of 5 μL of resazurin (25 μg mL^{-1}) and incubation for a further 3 hours at 37 °C in 5% CO_2. The fluorescence intensity was measured using a Tecan M1000 Pro monochromator plate reader, using automatic gain calculation CC_{50} were calculated by curve fitting the inhibition values against log(concentration) using sigmoidal dose-response function, with variable fitting values for bottom, top and slope. The maximal percentage of cytotoxicity is reported as D_{Max}, indicating any compounds with partial cytotoxicity. The curve fitting was implemented using Pipeline Pilot's dose-response component. Any value with > indicates a sample with no activity (low DMax value) or samples with CC_{50} values above the maximum tested concentration. Cytotoxic samples were classified by CC50 ≤ 32 μg mL^{-1} in either replicate.

8.17 Hydrolysis

Hydrolysis samples of complexes for investigation by NMR spectroscopy were prepared from a 4:1 mixture of d_3-acetonitrile/deuterium oxide to give a final concentration of 8 mg mL^{-1}. The NMR spectra of these samples were acquired every 24 hours over a five day period. Hydrolysis samples of complexes for investigation by UV/vis spectroscopy were prepared from a 4:1 mixture of acetonitrile/water to give a final concentration of 50 μM. The UV/vis spectra of these samples were

acquired every 24 hours over a five day period. After the five day period investigation period, the mass spectra of the hydrolysis samples were acquired.

8.18 Hydrophobicity

Equal volumes of 1-octanol and sodium chloride saturated distilled water were stirred for 16 hours and separated to give water-saturated octanol and octanol-saturated water. Standard solutions of each complex (5, 10, 20, 40 and 60 µM) were prepared in water-saturated octanol. The calibration curve of absorbance against concentration was determined from the maximum absorbance (λ_{max}) of the standard solutions. Stock solutions of each complex (50 µM) in water-saturated octanol (25 mL) were prepared. Six independent samples were prepared by the addition of the stock solution (3 mL) to a 15 mL Falcon tube followed by layered addition of octanol-saturated water (3 mL). Samples were shaken at 1000 g min^{-1} for 2 hours using an IKA Vibrax VXC basic shaker. The layers were separated and the water-saturated octanol layer retained. The concentration of each sample in the water-saturated octanol layer was determined by UV/vis spectroscopy with reference to the individual calibration curves to give an average concentration for shaken samples ([C]$_{final}$). The concentration of an unshaken sample of stock solution was determined to give [C]$_{initial}$. The partition coefficient (log P) was determined with **Equation 8.1**.

Equation 8.1: Determination of partition coefficient (log P)

$$\log P = \log_{10}\left(\frac{[C]_{final}}{[C]_{initial} - [C]_{final}}\right)$$

8.19 Biomembrane studies

The biomembrane studies were performed by Miss. Danielle Marriott and Dr. Shahrzad Mohamadi (University of Leeds).

The micro fabricated electrode coated with DOPC lipid was contained in a closed flow cell. A constant flow of phosphate-buffered saline (PBS) (pH 7.4) was passed over the electrode using a peristaltic pump at a flow rate of 5 - 10 mL min^{-1}. A constant flow of DOPC dispersion in PBS was deposited on the electrode with the

application of a potential excursion from -0.4 to -3.0 V at a scan rate of 100 Vs^{-1}. The electrode in the flow cell was connected to the PGSTATI2 potentiostat interfaced to a Powerlab signal generator and controlled by Scope software. A flow of argon gas is maintained over the electrolytes and the DOPC layer throughout. RCVs were obtained by applying a saw-tooth waveform from -0.4 to -1.2 V (vs Ag/AgCl) with ramp rate 40 V s^{-1} applied to the electrode surface. In the absence of faradaic reactions, the current on the RCV plot was directly proportional to the capacitance of the surface and is displayed as a function of voltage. All assays were carried out with 15.6 µM solutions of each complex in acetone with a constant flow of 0.1 M PBS. The complexes are sampled for 400 seconds followed by PBS for 400 seconds to allow *in situ* cleaning of the electrode.[24, 25]

8.20 References

1. J. Cosier and A. M. Glazer, *Journal of Applied Crystallography*, 1986, **19**, 105-107.

2. CrysAlisPRO, Oxford Diffraction / Agilent Technologies UK Ltd.

3. G. M. Sheldrick, *Crystallographic Computing 3: Data Collection, Structure Determination, Proteins, and Databases*, Clarendon Press Oxford, 1986.

4. G. M. Sheldrick, SHELXTL, 2014.

5. G. M. Sheldrick, *Acta Crystallographica A-Foundation and Advances*, 2015, **71**, 3-8.

6. G. M. Sheldrick, *Acta Crystallographica Section A*, 2008, **64**, 112-122.

7. O. V. Dolomanov, L. J. Bourhis, R. J. Gildea, J. A. K. Howard and H. Puschmann, *Journal of Applied Crystallography*, 2009, **42**, 339-341.

8. A. L. Spek, *Journal of Applied Crystallography*, 2003, **36**, 7-13.

9. Bruker AXS, DiffracEva Suite, 2009.

10. J. Laugier and B. Bochu, Chekcell, 2004.

11. C. F. Macrae, P. R. Edgington, P. McCabe, E. Pidcock, G. P. Shields, R. Taylor, M. Towler and J. van De Streek, *Journal of Applied Crystallography*, 2006, **39**, 453-457.

12. T. Cheng, W. Y. Yin, Y. Zhang, Y. N. Zhang and Y. Huang, *Org. Biomol. Chem.*, 2014, **12**, 1405-1411.

13. M. Colombo, S. Bossolo and A. Aramini, *J. Comb. Chem.*, 2009, **11**, 335-337.

14. D. W. Engers, C. M. Niswender, C. D. Weaver, S. Jadhav, U. N. Menon, R. Zamorano, P. J. Conn, C. W. Lindsley and C. R. Hopkins, *Journal of Medicinal Chemistry*, 2009, **52**, 4115-4118.

15. S. K. Kang, D. W. Kim and J. N. Park, *Synlett*, 2002, 427-430.

16. A. M. Martinez, N. Rodriguez, R. G. Arrayas and J. C. Carretero, *Chemical Communications*, 2014, **50**, 2801-2803.

17. C. Sambiagio, R. H. Munday, S. P. Marsden, A. J. Blacker and P. C. McGowan, *Chem.-Eur. J.*, 2014, **20**, 17606-17615.

18. S. Dutta, S. Pal and P. K. Bhattacharya, *Polyhedron*, 1999, **18**, 2157-2162.

19. L. Huang, Q. Li, C. Wang and C. Qi, *Journal of Organic Chemistry*, 2013, **78**, 3030-3038.

20.   E. V. Borisov, E. V. Skorodumov, V. M. Pachevskaya and P. E. Hansen, *Magnetic Resonance in Chemistry*, 2005, **43**, 992-998.

21.   X. Q. Han and R. A. Widenhoefer, *Journal of Organic Chemistry*, 2004, **69**, 1738-1740.

22.   L. Thomas, A. Gupta and V. Gupta, *Heterocyclic Communications*, 2002, **8**, 169-172.

23.   J. J. Hollick, B. T. Golding, I. R. Hardcastle, N. Martin, C. Richardson, L. J. M. Rigoreau, G. C. M. Smith and R. J. Griffin, *Bioorganic & Medicinal Chemistry Letters*, 2003, **13**, 3083-3086.

24.   Z. Coldrick, P. Steenson, P. Millner, M. Davies and A. Nelson, *Electrochimica Acta*, 2009, **54**, 4954-4962.

25.   S. Mohamadi, D. J. Tate, A. Vakurov and A. Nelson, *Analytica Chimica Acta*, 2014, **813**, 83-89.

Appendix

Crystallographic data

Complex	1.22	2.1a
Empirical formula	$C_{20}H_{14}N_2O$	$CoC_{37.33}H_{29}N_{6.67}O_3$
Formula weight	298.33	677.94
Size / mm³	0.35 × 0.11 × 0.06	0.19 × 0.13 × 0.06
Temperature / K	120.01(10)	121(1)
Wavelength	0.71073 (Mo-K_α)	0.71073 (Mo-K_α)
Crystal system	monoclinic	triclinic
Space group	$P\,2_1$	$P\,\overline{1}$
a / Å	10.8320(15)	9.9033(4)
b / Å	5.9238(7)	20.5749(8)
c / Å	11.2014(19)	24.2666(11)
α / °	90.00	81.265(4)
β / °	99.771(14)	80.376(4)
γ / °	90.00	80.584(3)
Volume / Å³	708.32(18)	4769.7(3)
Z	2	6
μ / mm⁻¹	0.088	0.589
F(000)	312.0	2104.0
Data collection range / °	7.38 ≤ θ ≤ 56.56	5.78 ≤ θ ≤ 52.74
Index ranges	-11 ≤ h ≤ 14	-12 ≤ h ≤ 12
	-7 ≤ k ≤ 7	-25 ≤ k ≤ 25
	-14 ≤ l ≤ 13	-30 ≤ l ≤ 26
Reflections collected	4234	68202
Independent reflections	3003 [R_{int} = 0.0472]	19506 [R_{int} = 0.1117]
Goodness of fit	1.165	1.117
Final R indices [I>2σ (I)]	R_1 = 0.0891, wR_2 = 0.1922	R_1 = 0.0958, wR_2 = 0.1717
R indices (all data)	R_1 = 0.1095, wR_2 = 0.2037	R_1 = 0.1493, wR_2 = 0.1935
Largest diff. / e Å⁻³	0.35 and -0.33	1.31 and -0.62

Complex	2.2	2.3
Empirical formula	$CoC_{36}H_{26}F_3N_6O_4$	$CoC_{36}H_{24}N_6O_3F_3$
Formula weight	722.56	704.54
Size / mm^3	0.31 × 0.28 × 0.18	0.14 × 0.04 × 0.02
Temperature / K	120.00(10)	120.00(14)
Wavelength	0.71073 (Mo-K_α)	1.54184 (Cu-K_α)
Crystal system	monoclinic	monoclinic
Space group	$P\,2_1/c$	$P\,2_1/n$
a / Å	9.4536(3)	17.142(3)
b / Å	14.7729(5)	9.9817(16)
c / Å	22.8060(9)	20.332(5)
α / °	90.00	90.00
β / °	97.774(3)	107.39(2)
γ / °	90.00	90.00
Volume / $Å^3$	3155.8(2)	3319.9(13)
Z	4	4
μ / mm^{-1}	0.614	4.590
F(000)	1480.0	1440.0
Data collection range / °	$5.802 \leq \theta \leq 62.478$	$5.936 \leq \theta \leq 150.194$
Index ranges	$-13 \leq h \leq 12$	$-17 \leq h \leq 20$
	$-19 \leq k \leq 21$	$-10 \leq k \leq 12$
	$-31 \leq l \leq 29$	$-25 \leq l \leq 24$
Reflections collected	21520	13790
Independent reflections	8980 [$R_{int} = 0.0377$]	6520 [$R_{int} = 0.0957$]
Goodness of fit	1.024	1.398
Final R indices [$I>2\sigma\,(I)$]	$R_1 = 0.0526$, $wR_2 = 0.1246$	$R_1 = 0.1569$, $wR_2 = 0.4001$
R indices (all data)	$R_1 = 0.0723$, $wR_2 = 0.1368$	$R_1 = 0.2005$, $wR_2 = 0.4365$
Largest diff. / e $Å^{-3}$	1.11 and -0.51	1.89 to -2.06

Complex	2.4	2.5
Empirical formula	$CoC_{36}H_{24}F_3N_6O_3$	$CoC_{38}H_{26.75}N_6O_{3.88}F_6$
Formula weight	704.54	802.33
Size / mm^3	$0.18 \times 0.09 \times 0.056$	$0.22 \times 0.07 \times 0.03$
Temperature / K	120.01(12)	120.00(17)
Wavelength	0.71073 (Mo-K_α)	0.71073 (Mo-K_α)
Crystal system	triclinic	triclinic
Space group	$P\overline{1}$	$P\overline{1}$
a / Å	10.2432(3)	10.1276(4)
b / Å	17.4043(6)	17.6691(8)
c / Å	19.9290(6)	20.7763(8)
α / °	105.613(3)	104.972(4)
β / °	98.051(2)	103.367(4)
γ / °	101.322(3)	97.194(4)
Volume / $Å^3$	3284.44(18)	3426.0(3)
Z	4	4
µ / mm^{-1}	0.586	0.586
F(000)	1440.0	1635.0
Data collection range / °	$6.154 \leq \theta \leq 62.508$	$6.08 \leq \theta \leq 62.44$
Index ranges	$-14 \leq h \leq 14$	$-13 \leq h \leq 14$
	$-23 \leq k \leq 23$	$-22 \leq k \leq 25$
	$-29 \leq l \leq 25$	$-30 \leq l \leq 26$
Reflections collected	48128	56214
Independent reflections	18850 [R_{int} = 0.0503]	19751 [R_{int} = 0.0967]
Goodness of fit	1.026	1.038
Final R indices [I>2σ (I)]	R_1 = 0.0601, wR_2 = 0.1078	R_1 = 0.0947, wR_2 = 0.1600
R indices (all data)	R_1 = 0.0938, wR_2 = 0.1204	R_1 = 0.1961, wR_2 = 0.2000
Largest diff. / e $Å^{-3}$	0.41 to -0.51	0.72 and -0.53

Complex	2.6	2.7
Empirical formula	$CoC_{36}H_{21}N_6O_3F_6$	$CoC_{39}H_{36}N_6O_6Cl_3$
Formula weight	758.52	850.02
Size / mm³	0.34 × 0.11 × 0.08	0.25 × 0.09 × 0.047
Temperature / K	120.01(10)	120.01(12)
Wavelength	0.71073 (Mo-K$_\alpha$)	0.71073 (Mo-K$_\alpha$)
Crystal system	monoclinic	monoclinic
Space group	$P\ 2_1/n$	$P\ 2_1/c$
a / Å	17.2118(8)	10.1077(5)
b / Å	9.8701(4)	24.5768(12)
c / Å	21.7012(13)	15.3694(7)
α / °	90.00	90.00
β / °	111.685(6)	95.048(4)
γ / °	90.00	90.00
Volume / Å³	3425.7(3)	3803.2(3)
Z	4	4
µ / mm^{-1}	0.579	0.718
F(000)	1536.0	1752.0
Data collection range / °	$6.282 \leq \theta \leq 62.394$	$6.27 \leq \theta \leq 52.744$
Index ranges	$-25 \leq h \leq 22$	$-12 \leq h \leq 10$
	$-13 \leq k \leq 13$	$-30 \leq k \leq 30$
	$-30 \leq l \leq 29$	$-14 \leq l \leq 19$
Reflections collected	28556	25381
Independent reflections	9755 [R_{int} = 0.0622]	7761 [R_{int} = 0.0540]
Goodness of fit	1.013	1.037
Final R indices [I>2σ (I)]	R_1 = 0.0560, wR_2 = 0.1250	R_1 = 0.0725, wR_2 = 0.2423
R indices (all data)	R_1 = 0.0975, wR_2 = 0.1430	R_1 = 0.0982, wR_2 = 0.2698
Largest diff. / e Å^{-3}	1.33 and -0.60	1.88 and -0.98

Complex	2.8	2.9
Empirical formula	$CoC_{41.6}H_{35.6}N_6O_3Cl_9$	$CoC_{37}H_{28}N_6O_4Br_3$
Formula weight	1045.54	919.31
Size / mm³	0.18 × 0.11 × 0.07	0.21 × 0.15 × 0.09
Temperature / K	110.01(10)	110.01(10)
Wavelength	0.71073 (Mo-K$_\alpha$)	0.71073 (Mo-K$_\alpha$)
Crystal system	triclinic	triclinic
Space group	$P\overline{1}$	$P\overline{1}$
a / Å	12.6110(10)	9.4660(4)
b / Å	13.4455(10)	11.0078(6)
c / Å	15.2414(13)	18.8301(10)
α / °	99.379(7)	102.546(5)
β / °	98.733(7)	93.541(4)
γ / °	115.762(8)	112.143(4)
Volume / Å³	2223.1(3)	1751.46(16)
Z	2	2
μ / mm⁻¹	0.974	3.962
F(000)	1062.0	912.0
Data collection range / °	$6.396 \leq \theta \leq 52.742$	$5.966 \leq \theta \leq 62.604$
Index ranges	$-14 \leq h \leq 15$	$-11 \leq h \leq 13$
	$-16 \leq k \leq 16$	$-15 \leq k \leq 11$
	$-19 \leq l \leq 19$	$-26 \leq l \leq 26$
Reflections collected	23994	21757
Independent reflections	9084 [R_{int} = 0.1005]	9870 [R_{int} = 0.0517]
Goodness of fit	1.015	1.046
Final R indices [I>2σ (I)]	R_1 = 0.0824, wR_2 = 0.1892	R_1 = 0.0629, wR_2 = 0.1476
R indices (all data)	R_1 = 0.1458, wR_2 = 0.2304	R_1 = 0.0908, wR_2 = 0.1647
Largest diff. / e Å⁻³	1.37 and -0.82	1.44 and -1.04

Complex	2.10	2.11
Empirical formula	$CoC_{37}H_{30}N_6O_5Br_3$	$CoC_{41}H_{26}N_6O_3F_9Cl_6$
Formula weight	937.33	1093.31
Size / mm³	$0.27 \times 0.21 \times 0.12$	$0.17 \times 0.04 \times 0.02$
Temperature / K	120.01(10)	120.02(17)
Wavelength	0.71073 (Mo-K$_\alpha$)	0.71073 (Mo-K$_\alpha$)
Crystal system	monoclinic	triclinic
Space group	$P\,2_1/c$	$P\,\overline{1}$
a / Å	16.3898(5)	10.8889(4)
b / Å	12.4501(4)	13.3504(8)
c / Å	18.6887(6)	17.0160(12)
α / °	90.00	67.229(6)
β / °	109.585(3)	75.622(5)
γ / °	90.00	76.343(4)
Volume / Å³	3592.9(2)	2182.1(2)
Z	4	2
μ / mm⁻¹	3.867	0.847
F(000)	1864.0	1096.0
Data collection range / °	$5.734 \leq \theta \leq 62.638$	$5.54 \leq \theta \leq 56.56$
Index ranges	$-23 \leq h \leq 23$	$-14 \leq h \leq 14$
	$-18 \leq k \leq 14$	$-17 \leq k \leq 17$
	$-26 \leq l \leq 24$	$-22 \leq l \leq 22$
Reflections collected	28543	30613
Independent reflections	10300 [R_{int} = 0.0425]	10824 [R_{int} = 0.0512]
Goodness of fit	1.038	1.035
Final R indices [I>2σ (I)]	R_1 = 0.0438, wR_2 = 0.0816	R_1 = 0.0707, wR_2 = 0.1585
R indices (all data)	R_1 = 0.0739, wR_2 = 0.0919	R_1 = 0.1035, wR_2 = 0.1771
Largest diff. / e Å⁻³	1.00 and -0.92	1.61 and -1.61

Complex	2.12	2.13
Empirical formula	$CoC_{41.25}H_{29.5}N_6O_{3.5}F_9Cl_{0.5}$	$CoC_{43}H_{22}N_6O_3F_{18}Cl_3$
Formula weight	912.86	1177.94
Size / mm³	0.28 × 0.05 × 0.02	0.18 × 0.06 × 0.03
Temperature / K	120.00(14)	120.01(19)
Wavelength	1.54184 (Cu-K$_\alpha$)	0.71073 (Mo-K$_\alpha$)
Crystal system	triclinic	monoclinic
Space group	$P\,\overline{1}$	$P\,2_1/c$
a / Å	14.3515(5)	15.525(2)
b / Å	16.6340(6)	16.581(2)
c / Å	18.2610(7)	18.791(3)
α / °	104.315(3)	90.00
β / °	98.187(3)	98.803(14)
γ / °	108.532(3)	90.00
Volume / Å³	3887.3(3)	4780.2(11)
Z	4	4
μ / mm^{-1}	4.613	0.644
F(000)	1854.0	2344.0
Data collection range / °	$6.508 \leq \theta \leq 147.904$	$5.852 \leq \theta \leq 52.848$
Index ranges	$-17 \leq h \leq 17$	$-19 \leq h \leq 19$
	$-13 \leq k \leq 19$	$-20 \leq k \leq 19$
	$-22 \leq l \leq 21$	$-23 \leq l \leq 23$
Reflections collected	32517	17371
Independent reflections	14768 [R_{int} = 0.0771]	17371 [R_{int}= 0.168]
Goodness of fit	1.023	0.865
Final R indices [I>2σ (I)]	R_1 = 0.0935, wR_2 = 0.2374	R_1 = 0.0933, wR_2 = 0.2084
R indices (all data)	R_1 = 0.1510, wR_2 = 0.2862	R_1 = 0.2201, wR_2 = 0.2425
Largest diff. / e Å^{-3}	1.56 and -1.02	0.96 and -0.84

Complex	2.14a	2.15a
Empirical formula	$CoC_{40}H_{38}N_6O_{4.5}$	$CoC_{41.91}H_{44.66}N_6O_{5.92}$
Formula weight	733.69	786.05
Size / mm³	$0.27 \times 0.19 \times 0.09$	$0.18 \times 0.10 \times 0.09$
Temperature / K	120.01(10)	120.0(3)
Wavelength	0.71073 (Mo-K$_\alpha$)	0.71073 (Mo-K$_\alpha$)
Crystal system	triclinic	triclinic
Space group	$P\overline{1}$	$P\overline{1}$
a / Å	9.5494(3)	11.6389(4)
b / Å	13.7457(5)	13.5700(6)
c / Å	15.0145(5)	14.0659(7)
α / °	78.179(3)	69.506(4)
β / °	73.207(3)	72.633(4)
γ / °	70.571(3)	83.743(3)
Volume / Å³	1766.40(12)	1986.08(16)
Z	2	2
μ / mm^{-1}	0.539	0.486
F(000)	766.0	825.0
Data collection range / °	$6.186 \le \theta \le 62.478$	$6.136 \le \theta \le 62.426$
Index ranges	$-13 \le h \le 13$	$-16 \le h \le 15$
	$-19 \le k \le 18$	$-19 \le k \le 18$
	$-21 \le l \le 21$	$-18 \le l \le 20$
Reflections collected	29518	25169
Independent reflections	10124 [R_{int} = 0.0412]	11192 [R_{int} = 0.0401]
Goodness of fit	1.048	1.058
Final R indices [I>2σ (I)]	R_1 = 0.0470, wR_2 = 0.0993	R_1 = 0.0571, wR_2 = 0.1302
R indices (all data)	R_1 = 0.0609, wR_2 = 0.1064	R_1 = 0.0794, wR_2 = 0.1419
Largest diff. / e Å^{-3}	0.47 and -0.47	1.06 to -0.47

Complex	2.16a	2.17a
Empirical formula	$CoC_{46.13}H_{46.75}N_6O_{3.25}Cl_{6.75}$	$CoC_{40}H_{33}N_6O_7$
Formula weight	1035.36	768.65
Size / mm³	0.37 × 0.14 × 0.04	0.25 × 0.08 × 0.06
Temperature / K	120.02(16)	120.01(18)
Wavelength	0.71073 (Mo-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	triclinic	triclinic
Space group	$P\overline{1}$	$P\overline{1}$
a / Å	13.7483(6)	10.0253(4)
b / Å	17.0078(6)	13.0399(5)
c / Å	23.1559(9)	14.7478(5)
α / °	78.587(3)	98.078(3)
β / °	82.526(4)	108.411(4)
γ / °	70.642(4)	105.152(4)
Volume / Å³	4995.2(4)	1712.53(12)
Z	4	2
μ / mm⁻¹	0.750	4.459
F(000)	2133.0	796.0
Data collection range / °	5.772 ≤ θ ≤ 46.512	6.52 ≤ θ ≤ 147.46
Index ranges	-15 ≤ h ≤ 15	-12 ≤ h ≤ 11
	-18 ≤ k ≤ 18	-16 ≤ k ≤ 15
	-25 ≤ l ≤ 25	-18 ≤ l ≤ 15
Reflections collected	61654	13889
Independent reflections	14319 [R_{int} = 0.1000]	6471 [R_{int} = 0.0295]
Goodness of fit	1.017	1.032
Final R indices [I>2σ (I)]	R_1 = 0.1076, wR_2 = 0.2892	R_1 = 0.0462, wR_2 = 0.1207
R indices (all data)	R_1 = 0.1546, wR_2 = 0.3320	R_1 = 0.0513, wR_2 = 0.1253
Largest diff. / e Å⁻³	2.29 to -1.25	0.99 and -1.43

Complex	2.1b	2.14b
Empirical formula	$CoC_{36}H_{29}N_6O_4$	$CoC_{40}H_{44}N_6O_6$
Formula weight	668.58	763.74
Size / mm³	0.11 × 0.08 × 0.05	0.14 × 0.11 × 0.04
Temperature / K	120.00(13)	120.0(2)
Wavelength	0.71073 (Mo-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	triclinic	triclinic
Space group	$P\,\overline{1}$	$P\,\overline{1}$
a / Å	11.5229(13)	14.7347(4)
b / Å	11.9759(13)	15.5381(4)
c / Å	12.0279(8)	18.5489(5)
α / °	78.077(7)	111.770(2)
β / °	78.949(7)	93.953(2)
γ / °	77.062(9)	106.249(2)
Volume / Å³	1564.3(3)	3714.61(15)
Z	2	4
μ / mm^{-1}	0.600	4.080
F(000)	692.0	1604.0
Data collection range / °	6.44 ≤ θ ≤ 52.74	6.36 ≤ θ ≤ 147.82
Index ranges	-14 ≤ h ≤ 14	-18 ≤ h ≤ 14
	-14 ≤ k ≤ 14	-19 ≤ k ≤ 17
	-15 ≤ l ≤ 13	-23 ≤ l ≤ 22
Reflections collected	16177	34545
Independent reflections	6368 [R_{int} = 0.0582]	14098 [R_{int} = 0.0447]
Goodness of fit	1.047	1.044
Final R indices [I>2σ (I)]	R_1 = 0.0512, wR_2 = 0.1015	R_1 = 0.0765, wR_2 = 0.2180
R indices (all data)	R_1 = 0.0677, wR_2 = 0.1096	R_1 = 0.0988, wR_2 = 0.2400
Largest diff. / e Å^{-3}	0.70 and -0.38	1.74 and -0.84

Complex	2.15b	2.16b
Empirical formula	$CoC_{40.85}H_{36.85}N_5O_4Cl_{2.55}$	$CoC_{42}H_{51.67}N_6O_{6.33}$
Formula weight	811.13	800.82
Size / mm³	0.19 × 0.12 × 0.06	0.09 × 0.05 × 0.04
Temperature / K	120.1(4)	120.5(9)
Wavelength	0.71073 (Mo-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	triclinic	triclinic
Space group	$P\,\overline{1}$	$P\,\overline{1}$
a / Å	11.5554(11)	11.0281(6)
b / Å	12.0050(10)	13.7282(5)
c / Å	15.0864(15)	14.9976(7)
α / °	95.392(7)	66.892(4)
β / °	100.331(8)	74.161(4)
γ / °	99.212(8)	77.568(4)
Volume / Å³	2016.1(3)	1994.03(16)
Z	2	2
μ / mm^{-1}	0.641	0.486
F(000)	839.0	847.0
Data collection range / °	6.148 ≤ θ ≤ 53.006	3.02 ≤ θ ≤ 50.24
Index ranges	-14 ≤ h ≤ 13	-13 ≤ h ≤ 10
	-14 ≤ k ≤ 14	-15 ≤ k ≤ 16
	-18 ≤ l ≤ 18	-16 ≤ l ≤ 17
Reflections collected	14935	14967
Independent reflections	14935 [R_{int} = 0.152]	6863 [R_{int} = 0.044]
Goodness of fit	0.985	1.082
Final R indices [I>2σ (I)]	R_1 = 0.1253, wR_2 = 0.3307	R_1 = 0.0889, wR_2 = 0.2356
R indices (all data)	R_1 = 0.2379, wR_2 = 0.3762	R_1 = 0.1076, wR_2 = 0.2493
Largest diff. / e Å^{-3}	1.41 and -0.84	1.16 and -0.97

Complex	2.17b	2.18b
Empirical formula	$CoC_{39}H_{35}N_6O_7$	$CoC_{42}H_{50}N_6O_{14}$
Formula weight	758.66	921.81
Size / mm^3	$0.13 \times 0.07 \times 0.06$	$0.15 \times 0.13 \times 0.11$
Temperature / K	120.01(16)	120.0(2)
Wavelength	1.54184 (Cu-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	triclinic	triclinic
Space group	$P\,\overline{1}$	$P\,\overline{1}$
a / Å	10.5395(4)	13.8190(3)
b / Å	13.1143(5)	15.3377(4)
c / Å	13.5741(5)	22.5261(6)
α / °	79.375(3)	76.712(2)
β / °	69.846(3)	75.834(2)
γ / °	85.467(3)	67.105(2)
Volume / Å^3	1730.87(11)	4215.83(19)
Z	2	4
µ / mm^{-1}	4.402	3.843
F(000)	788.0	1932.0
Data collection range / °	$6.86 \leq \theta \leq 147.22$	$6.32 \leq \theta \leq 147.62$
Index ranges	$-13 \leq h \leq 9$	$-17 \leq h \leq 16$
	$-16 \leq k \leq 14$	$-18 \leq k \leq 18$
	$-16 \leq l \leq 15$	$-27 \leq l \leq 27$
Reflections collected	13294	38898
Independent reflections	6533 [R_{int} = 0.0329]	15936 [R_{int} = 0.0319]
Goodness of fit	1.019	1.023
Final R indices [I>2σ (I)]	R_1 = 0.0475, wR_2 = 0.1137	R_1 = 0.0437, wR_2 = 0.1127
R indices (all data)	R_1 = 0.0592, wR_2 = 0.1226	R_1 = 0.0570, wR_2 = 0.1210
Largest diff. / e Å^{-3}	0.71 and -0.61	0.66 and -0.65

Complex	3.1	3.2
Empirical formula	$CoC_{26}H_{20}N_6O_2S_2$	$CoC_{28}H_{24}N_6O_2S_2$
Formula weight	571.53	599.58
Size / mm³	$0.21 \times 0.15 \times 0.11$	$0.13 \times 0.11 \times 0.04$
Temperature / K	120.00(10)	120.00(13)
Wavelength	0.71073 (Mo-K_α)	0.71073 (Mo-K_α)
Crystal system	triclinic	monoclinic
Space group	$P\overline{1}$	$P\,2_1/c$
a / Å	8.1387(6)	8.0460(9)
b / Å	8.7460(5)	21.181(2)
c / Å	9.8413(7)	8.9024(11)
α / °	85.840(6)	90.00
β / °	75.883(6)	113.347(14)
γ / °	66.742(7)	90.00
Volume / Å³	623.93(8)	1392.9(3)
Z	1	2
μ / mm⁻¹	0.892	0.803
F(000)	293.0	618.0
Data collection range / °	$6.544 \leq \theta \leq 56.556$	$5.84 \leq \theta \leq 62.5$
Index ranges	$-10 \leq h \leq 10$	$-9 \leq h \leq 11$
	$-11 \leq k \leq 11$	$-28 \leq k \leq 29$
	$-13 \leq l \leq 13$	$-12 \leq l \leq 7$
Reflections collected	1243	9421
Independent reflections	3094 [R_{int} = 0.0458]	3988 [R_{int} = 0.0477]
Goodness of fit	1.077	1.059
Final R indices [I>2σ (I)]	R_1 = 0.0354, wR_2 = 0.0789	R_1 = 0.0544, wR_2 = 0.1123
R indices (all data)	R_1 = 0.0430, wR_2 = 0.0838	R_1 = 0.0929, wR_2 = 0.1302
Largest diff. / e Å⁻³	0.36 and -0.29	0.66 and -0.58

Complex	3.3	3.4
Empirical formula	$CoC_{30}H_{28}N_6O_2S_2$	$CoC_{28}H_{24}N_6O_2S_2$
Formula weight	627.63	599.58
Size / mm³	0.2 × 0.14 × 0.09	0.16 × 0.12 × 0.08
Temperature / K	120.01(10)	119.99(17)
Wavelength	pink block	0.71073 (Mo-K$_\alpha$)
Crystal system	triclinic	monoclinic
Space group	$P\,\overline{1}$	$P\,2_1/c$
a / Å	7.8650(8)	10.4338(14)
b / Å	8.4741(8)	13.6460(14)
c / Å	12.8131(13)	19.816(3)
α / °	105.541(9)	90.00
β / °	91.317(8)	103.722(16)
γ / °	116.677(10)	90.00
Volume / Å³	724.92(12)	2740.9(6)
Z	1	4
μ / mm⁻¹	0.775	0.816
F(000)	325.0	1236.0
Data collection range / °	6.26 ≤ θ ≤ 62.3	5.98 ≤ θ ≤ 62.46
Index ranges	-11 ≤ h ≤ 10	-15 ≤ h ≤ 10
	-11 ≤ k ≤ 12	-19 ≤ k ≤ 19
	-17 ≤ l ≤ 11	-26 ≤ l ≤ 28
Reflections collected	5365	21138
Independent reflections	3534 [R_{int} = 0.0333]	7795 [R_{int} = 0.0911]
Goodness of fit	1.061	1.087
Final R indices [I>2σ (I)]	R_1 = 0.0635, wR_2 = 0.1534	R_1 = 0.0840, wR_2 = 0.1878
R indices (all data)	R_1 = 0.0807, wR_2 = 0.1680	R_1 = 0.1317, wR_2 = 0.2179
Largest diff. / e Å⁻³	1.45 and -0.65	1.58 and -0.80

Complex	3.5	3.7
Empirical formula	$CoC_{28}H_{24}N_6O_2S_2$	$CoC_{28}H24N_6O_4S_2$
Formula weight	599.58	1263.16
Size / mm³	0.35 × 0.07 × 0.04	0.24 × 0.07 × 0.02
Temperature / K	120.0(2)	120.2(6)
Wavelength	0.71073 (Mo-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	monoclinic	monoclinic
Space group	C c	C c
a / Å	8.2408(7)	8.51272(16)
b / Å	20.8725(16)	20.4558(3)
c / Å	16.2881(13)	16.2386(3)
α / °	90.00	90.00
β / °	98.690(8)	97.6694(17)
γ / °	90.00	90.00
Volume / Å³	2769.5(4)	2802.40(9)
Z	4	2
μ / mm^{-1}	0.808	6.584
F(000)	1236.0	1300.0
Data collection range / °	$6.24 \leq \theta \leq 62.7$	$8.64 \leq \theta \leq 147.44$
Index ranges	$-8 \leq h \leq 11$	$-10 \leq h \leq 9$
	$-30 \leq k \leq 20$	$-24 \leq k \leq 17$
	$-23 \leq l \leq 18$	$-18 \leq l \leq 19$
Reflections collected	10913	5343
Independent reflections	5736 [R_{int} = 0.0497]	3493 [R_{int} = 0.0282]
Goodness of fit	1.039	1.036
Final R indices [I>2σ (I)]	R_1 = 0.0513, wR_2 = 0.0791	R_1 = 0.0298, wR_2 = 0.0767
R indices (all data)	R_1 = 0.0656, wR_2 = 0.0843	R_1 = 0.0312, wR_2 = 0.0777
Largest diff. / e Å^{-3}	0.81 and -0.46	0.32 and -0.37

Complex	3.8	3.9
Empirical formula	$CoC_{30}H_{28}N_6O_6S_2$	$CoC_{24}H_{20}N_4O_2Cl_2$
Formula weight	691.63	526.27
Size / mm³	0.31 × 0.14 × 0.10	0.13 × 0.11 × 0.09
Temperature / K	119.99(14)	120.01(120
Wavelength	0.71073 (Mo-K$_\alpha$)	0.71073 (Mo-K$_\alpha$)
Crystal system	triclinic	monoclinic
Space group	$P\,\overline{1}$	$P\,2_1/c$
a / Å	14.4852(5)	10.3023(12)
b / Å	15.8534(5)	12.9556(8)
c / Å	27.0438(8)	9.3713(10)
α / °	90.006(2)	90.00
β / °	90.054(3)	115.732(13)
γ / °	104.182(3)	90.00
Volume / Å³	6021.0(3)	1126.77(18)
Z	8	2
μ / mm⁻¹	0.764	1.029
F(000)	2856.0	538.0
Data collection range / °	5.59 ≤ θ ≤ 62.688	5.76 ≤ θ ≤ 62.42
Index ranges	-20 ≤ h ≤ 20	-15 ≤ h ≤ 11
	-22 ≤ k ≤ 23	-12 ≤ k ≤ 18
	-38 ≤ l ≤ 33	-9 ≤ l ≤ 13
Reflections collected	67052	6848
Independent reflections	31516 [R_{int} = 0.0525]	3193 [R_{int} = 0.0371]
Goodness of fit	1.005	1.059
Final R indices [I>2σ (I)]	R_1 = 0.0550, wR_2 = 0.1026	R_1 = 0.0500, wR_2 = 0.0995
R indices (all data)	R_1 = 0.0882, wR_2 = 0.1182	R_1 = 0.0692, wR_2 = 0.1075
Largest diff. / e Å⁻³	0.74 and -0.69	0.93 and -0.38

Complex	3.10	4.1
Empirical formula	$CoC_{24}H_{24}N_4O_4I_2$	$CoC_{33}H_{21}N_4O_4F_{12}$
Formula weight	745.20	824.47
Size / mm³	0.24 × 0.16 × 0.07	0.08 × 0.05 × 0.02
Temperature / K	120.01(10)	120.3(7)
Wavelength	0.71073 (Mo-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	monoclininc	tetragonal
Space group	$P\,2_1/c$	$I\,4_1cd$
a / Å	8.2666(2)	19.8988(6)
b / Å	11.2691(3)	19.8988(6)
c / Å	14.7572(5)	17.2717(11)
α / °	90.00	90.00
β / °	105.541(3)	90.00
γ / °	90.00	90.00
Volume / Å³	1324.49(7)	6838.9(6)
Z	2	8
μ / mm⁻¹	3.015	4.944
F(000)	722.0	3312.0
Data collection range / °	$6.26 \leq \theta \leq 62.4$	$8.888 \leq \theta \leq 147.232$
Index ranges	$-8 \leq h \leq 11$	$-15 \leq h \leq 21$
	$-15 \leq k \leq 16$	$-21 \leq k \leq 24$
	$-19 \leq l \leq 21$	$-20 \leq l \leq 20$
Reflections collected	11519	7432
Independent reflections	3880 [R_{int} = 0.0552]	2869 [R_{int} = 0.0694]
Goodness of fit	1.070	1.025
Final R indices [I>2σ (I)]	R_1 = 0.0408, wR_2 = 0.0826	R_1 = 0.0597, wR_2 = 0.1411
R indices (all data)	R_1 = 0.0542, wR_2 = 0.0952	R_1 = 0.0822, wR_2 = 0.1548
Largest diff. / e Å⁻³	14.4 to -1.44	0.55 and -0.40

Complex	4.2	4.3
Empirical formula	$CoC_{43}H_{0.5}N_4O_4F_{12}$	$C_{38}H_{23}CoN_4O_4F_{12}$
Formula weight	923.90	886.53
Size / mm³	0.26 × 0.06 × 0.04	0.19 × 0.09 × 0.01
Temperature / K	120.0(2)	120.0(2)
Wavelength	1.54184 (Cu-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	triclinic	monoclinic
Space group	$P\ \overline{1}$	$P\ 2_1/n$
a / Å	10.126(3)	20.8681(5)
b / Å	10.133(3)	15.8504(4)
c / Å	23.332(3)	22.6017(5)
α / °	78.006(16)	90.00
β / °	77.972(16)	99.010(2)
γ / °	60.60(3)	90.00
Volume / Å³	2024.0(9)	7383.7(3)
Z	2	8
μ / mm⁻¹	4.264	4.628
F(000)	907.0	3568.0
Data collection range / °	7.808 ≤ θ ≤ 148.966	6.276 ≤ θ ≤ 148.944
Index ranges	-11 ≤ h ≤ 12	-25 ≤ h ≤ 26
	-8 ≤ k ≤ 12	-14 ≤ k ≤ 19
	-28 ≤ l ≤ 25	-25 ≤ l ≤ 28
Reflections collected	8696	41615
Independent reflections	5795 [R_{int} = 0.0748]	14917 [R_{int} = 0.1009]
Goodness of fit	2.106	1.017
Final R indices [I>2σ (I)]	R_1 = 0.2377, wR_2 = 0.5589	R_1 = 0.0687, wR_2 = 0.1503
R indices (all data)	R_1 = 0.2586, wR_2 = 0.5714	R_1 = 0.1155, wR_2 = 0.1777
Largest diff. / e Å⁻³	2.24 and -0.98	0.73 and -0.54

Complex	4.4	4.6
Empirical formula	$CoC_{38}H_{22}N_4O_4F_{13}$	$CoC_{39}H_{22}N_4O_4F_{15}$
Formula weight	904.53	954.54
Size / mm³	0.28 × 0.19 × 0.04	0.21 × 0.19 × 0.09
Temperature / K	120.00(15)	119.99(14)
Wavelength	1.54184 (Cu-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	monoclinic	monoclinic
Space group	$P\,2_1/c$	$P\,2_1/c$
a / Å	13.18694(13)	13.37591(16)
b / Å	15.53624(17)	15.71209(18)
c / Å	18.1682(2)	18.3039(2)
α / °	90.00	90.00
β / °	96.7273(10)	98.2790(12)
γ / °	90.00	90.00
Volume / Å³	3696.59(7)	3806.72(8)
Z	4	4
μ / mm⁻¹	4.676	4.655
F(000)	1816.0	1912.0
Data collection range / °	$6.74 \leq \theta \leq 147.48$	$6.68 \leq \theta \leq 147.42$
Index ranges	$-16 \leq h \leq 12$	$-11 \leq h \leq 16$
	$-12 \leq k \leq 18$	$-16 \leq k \leq 19$
	$-21 \leq l \leq 22$	$-22 \leq l \leq 21$
Reflections collected	15160	16206
Independent reflections	7268 [R_{int} = 0.0294]	7489 [R_{int} = 0.0369]
Goodness of fit	1.015	1.016
Final R indices [I>2σ (I)]	R_1 = 0.0365, wR_2 = 0.0882	R_1 = 0.0398, wR_2 = 0.0936
R indices (all data)	R_1 = 0.0441, wR_2 = 0.0935	R_1 = 0.0490, wR_2 = 0.0993
Largest diff. / e Å⁻³	0.41 and -0.34	0.39 and -0.30

Complex	4.8	4.9
Empirical formula	$CoC_{40}H_{27}N_4O_4F_{12}Cl_2$	$CoC_{39.6}H_{26.2}N_4O_5F_{12}Cl_{1.2}$
Formula weight	985.48	967.51
Size / mm³	0.33 × 0.29 × 0.08	0.18 × 0.11 × 0.05
Temperature / K	120.0(2)	120.01(19)
Wavelength	1.54184 (Cu-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	monoclinic	monoclinic
Space group	C c	C c
a / Å	18.9964(3)	19.123(2)
b / Å	9.37144(13)	9.3137(11)
c / Å	22.8371(3)	22.8702(16)
α / °	90.00	90.00
β / °	96.2779(13)	96.100(9)
γ / °	90.00	90.00
Volume / Å³	4041.16(10)	4050.2(7)
Z	4	4
μ / mm^{-1}	5.481	5.003
F(000)	1984.0	1949.0
Data collection range / °	7.79 ≤ θ ≤ 148.85	7.776 ≤ θ ≤ 147.17
Index ranges	-22 ≤ h ≤ 23	-23 ≤ h ≤ 20
	-11 ≤ k ≤ 11	-9 ≤ k ≤ 11
	-28 ≤ l ≤ 25	-23 ≤ l ≤ 28
Reflections collected	14844	8856
Independent reflections	5997 [R_{int} = 0.0643]	4807 [R_{int} = 0.0668]
Goodness of fit	1.060	1.029
Final R indices [I>2σ (I)]	R_1 = 0.0666, wR_2 = 0.1667	R_1 = 0.0997, wR_2 = 0.2631
R indices (all data)	R_1 = 0.0740, wR_2 = 0.1753	R_1 = 0.1289, wR_2 = 0.2972
Largest diff. / e Å^{-3}	0.78 and -0.60	1.08 and -0.50

Complex	4.11	4.13
Empirical formula	$CoC_{43}H_{25}N_4O_4F_{15}Cl_3Fe$	$Co_{0.13}C_{47}H_{29.7}N_4O_{4.35}F_{12}Fe_2$
Formula weight	1167.80	1067.11
Size / mm³	0.10 × 0.10 × 0.06	0.15 × 0.06 × 0.04
Temperature / K	119.99(11)	120.2(5)
Wavelength	1.54184 (Cu-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	orthorhombic	monoclinic
Space group	P na2$_1$	P 2$_1$/c
a / Å	15.6045(2)	18.0839(10)
b / Å	17.4409(2)	21.4884(16)
c / Å	16.7895(3)	21.4884(16)
α / °	90.00	90.00
β / °	90.00	99.405(6)
γ / °	90.00	90.00
Volume / Å³	4569.36(12)	8762.5(10)
Z	4	8
μ / mm^{-1}	7.999	6.588
F(000)	2328.0	4303.0
Data collection range / °	7.308 ≤ θ ≤ 147.612	6.44 ≤ θ ≤ 148.988
Index ranges	-18 ≤ h ≤ 17	-16 ≤ h ≤ 22
	-21 ≤ k ≤ 17	-21 ≤ k ≤ 26
	-19 ≤ l ≤ 20	-26 ≤ l ≤ 21
Reflections collected	11890	40239
Independent reflections	7128 [R$_{int}$ = 0.0330]	17035 [R$_{int}$ = 0.0991]
Goodness of fit	1.022	1.054
Final R indices [I>2σ (I)]	R$_1$ = 0.0368, wR$_2$ = 0.0836	R$_1$ = 0.1010, wR$_2$ = 0.2470
R indices (all data)	R$_1$ = 0.0431, wR$_2$ = 0.0874	R$_1$ = 0.1636, wR$_2$ = 0.2886
Largest diff. / e Å^{-3}	0.71 and -0.31	1.71 and -0.61

Complex	4.14	4.15
Empirical formula	$CoC_{47}H_{28}N_4O_4F_{13}Fe$	$CoC_{49}H_{30}N_4O_4F_{13}Cl_6Fe$
Formula weight	1074.51	1313.25
Size / mm^3	$0.13 \times 0.10 \times 0.09$	$0.18 \times 0.08 \times 0.07$
Temperature / K	119.99(19)	119.99(17)
Wavelength	1.54184 (Cu-K_α)	1.54184 (Cu-K_α)
Crystal system	monoclinic	monoclinic
Space group	P 2/c	I a
a / Å	9.5681(7)	23.2888(3)
b / Å	9.9559(6)	11.92619(14)
c / Å	23.4877(19)	19.5918(2)
α / °	90.00	90.00
β / °	98.410(8)	106.1697(13)
γ / °	90.00	90.00
Volume / $Å^3$	2213.4(3)	5226.30(11)
Z	2	4
μ / mm^{-1}	6.508	8.392
F(000)	1080.0	2624.0
Data collection range / °	$7.61 \leq \theta \leq 148.26$	$7.906 \leq \theta \leq 148.604$
Index ranges	$-11 \leq h \leq 11$	$-28 \leq h \leq 29$
	$-12 \leq k \leq 12$	$-14 \leq k \leq 14$
	$-29 \leq l \leq 24$	$-23 \leq l \leq 24$
Reflections collected	14668	19257
Independent reflections	4461 [R_{int} = 0.0548]	7484 [R_{int} = 0.0304]
Goodness of fit	1.078	1.030
Final R indices [I>2σ (I)]	R_1 = 0.1409, wR_2 = 0.3462	R_1 = 0.0725, wR_2 = 0.1965
R indices (all data)	R_1 = 0.1558, wR_2 = 0.3560	R_1 = 0.0779, wR_2 = 0.2039
Largest diff. / e $Å^{-3}$	0.96 and -0.86	1.81 and -0.95

Complex	4.16	4.17
Empirical formula	$CoC_{47}H_{27}N_4O_4F_{14}Fe$	$CoC_{50.8}H_{36.6}N_4O_{5.4}F_{12}Fe$
Formula weight	1092.50	1132.22
Size / mm³	0.35 × 0.07 × 0.06	0.20 × 0.19 × 0.15
Temperature / K	120.0(2)	120.00(15)
Wavelength	1.54184 (Cu-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	monoclinic	orthorhombic
Space group	$P\,2_1$	$P\,na2_1$
a / Å	9.7396(7)	9.8368(2)
b / Å	22.542(3)	21.8083(6)
c / Å	20.980(2)	22.8353(6)
α / °	90.00	90.00
β / °	92.779(8)	90.00
γ / °	90.00	90.00
Volume / Å³	4600.9(8)	4898.7(2)
Z	4	4
μ / mm^{-1}	6.305	5.904
F(000)	2192.0	2294.0
Data collection range / °	7.844 ≤ θ ≤ 101.442	7.744 ≤ θ ≤ 147.138
Index ranges	-9 ≤ h ≤ 9	-6 ≤ h ≤ 11
	-22 ≤ k ≤ 20	-14 ≤ k ≤ 26
	-15 ≤ l ≤ 20	-27 ≤ l ≤ 26
Reflections collected	12026	12661
Independent reflections	7818 [R_{int} = 0.1133]	7527 [R_{int} = 0.0442]
Goodness of fit	1.525	1.035
Final R indices [I>2σ (I)]	R_1 = 0.1644, wR_2 = 0.4012	R_1 = 0.0612, wR_2 = 0.1595
R indices (all data)	R_1 = 0.1812, wR_2 = 0.4203	R_1 = 0.0687, wR_2 = 0.1681
Largest diff. / e Å^{-3}	2.42 and -0.98	0.77 and -0.81

Complex	4.18	4.19
Empirical formula	$CoC_{48}H_{31}N_4O_4F_{12}Fe$	$CoC_{49.83}H_{37.33}N_4O_{5.33}F_{12}Fe$
Formula weight	1082.20	1120.26
Size / mm³	0.21 × 0.10 × 0.07	0.20 × 0.10 × 0.07
Temperature / K	120.01(16)	120.1(3)
Wavelength	1.54184 (Cu-K$_\alpha$)	1.54184 (Cu-K$_\alpha$)
Crystal system	monoclinic	orthorhombic
Space group	$P\,2_1/c$	$P\,na2_1$
a / Å	19.7402(4)	9.7226(4)
b / Å	10.0264(2)	21.5917(12)
c / Å	23.0424(5)	23.4965(12)
α / °	90.00	90.00
β / °	104.191(2)	90.00
γ / °	90.00	90.00
Volume / Å³	4421.45(16)	4932.5(4)
Z	4	4
μ / mm^{-1}	6.882	5.854
F(000)	2183.0	2272.0
Data collection range / °	7.916 ≤ θ ≤ 147.666	8.19 ≤ θ ≤ 147.416
Index ranges	-24 ≤ h ≤ 23	-11 ≤ h ≤ 8
	-12 ≤ k ≤ 8	-18 ≤ k ≤ 26
	-26 ≤ l ≤ 28	-28 ≤ l ≤ 26
Reflections collected	16774	12711
Independent reflections	8644 / 42 / 659	7418 [R_{int} = 0.0612]
Goodness of fit	1.012	1.035
Final R indices [I>2σ (I)]	R_1 = 0.0497, wR_2 = 0.1142	R_1 = 0.0853, wR_2 = 0.2102
R indices (all data)	R_1 = 0.0761, wR_2 = 0.1288	R_1 = 0.1152, wR_2 = 0.2341
Largest diff. / e Å^{-3}	0.59 and -0.60	0.74 and -0.63

Complex	Hydrolysis product
Empirical formula	$FeC_{42}H_{30}O_6F_9$
Formula weight	1025.06
Size / mm^3	0.08 × 0.04 × 0.02
Temperature / K	119.97(16)
Wavelength	1.54184 (Cu-K$_\alpha$)
Crystal system	monoclinic
Space group	$P\,2_1/c$
a / Å	10.7214(2)
b / Å	36.8049(10)
c / Å	10.5177(3)
α / °	90.00
β / °	95.471(2)
γ / °	90.00
Volume / Å^3	4131.35(18)
Z	4
μ / mm^{-1}	11.822
F(000)	2060.0
Data collection range / °	$8.284 \leq \theta \leq 147.53$
Index ranges	$-13 \leq h \leq 10$
	$-30 \leq k \leq 41$
	$-12 \leq l \leq 9$
Reflections collected	18613
Independent reflections	7844 [R_{int} = 0.0408]
Goodness of fit	1.030
Final R indices [I>2σ (I)]	R_1 = 0.0451, wR_2 = 0.0964
R indices (all data)	R_1 = 0.0640, wR_2 = 0.1044
Largest diff. / e Å^{-3}	0.51 and -0.44