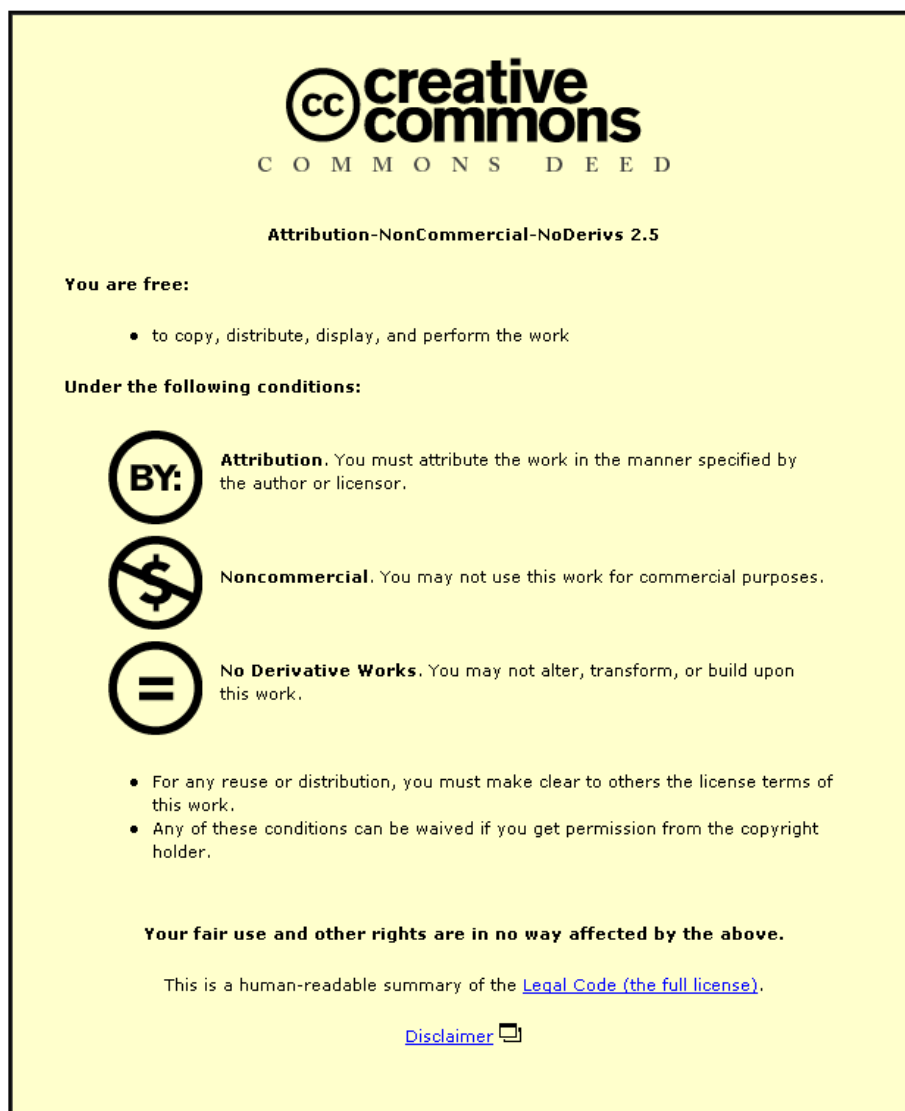


This item was submitted to Loughborough University as a PhD thesis by the author and is made available in the Institutional Repository (<https://dspace.lboro.ac.uk/>) under the following Creative Commons Licence conditions.



For the full text of this licence, please go to:
<http://creativecommons.org/licenses/by-nc-nd/2.5/>



Pilkington Library

Author/Filing Title WEIGHTMAN, J.S

Accession/Copy No. 060129861

Vol. No. Class Mark

27 JUN 1997

26 JUN 1998

25 JUN 1999

LOAN COPY

0401298612



BADMINTON PRESS
18 THE HALFCROFT
SYSTON
LEICESTER LE7 4LD
ENGLAND
TEL : 0116 260 2917
FAX : 0116 269 6639

Spectroscopic and Electrochemical Sensing of Anions and Cations
Using Novel Receptor Molecules

by

John S. Weightman

A Doctoral Thesis

Submitted in partial fulfilment of the requirements

for the award of

Degree of Doctor of Philosophy of Loughborough University

30/9/96

© by John S. Weightman

 Loughborough University	
Doc	M-97
Class	
Acc	
No	040129861

99098199

Contents

	Page No.
Abstract	(i)
Acknowledgements	(ii)
Chapter 1 - Introduction	1
1.0 Introduction	1
1.1 Biotic (natural) and abiotic (synthetic) receptors	6
1.2 Abiotic receptors	9
1.2.1 Receptors for cationic guest species	9
1.2.2 Receptors for neutral guest species	14
1.2.3 Receptors for anionic guest species	18
1.3 Abiotic host molecules that contain a responsive function	35
1.3.1 Electrochemical recognition of cations	36
1.3.2 Electrochemical recognition of neutral guest species	40
1.3.3 Electrochemical recognition of anions	42
1.3.4 Photoresponsive - Fluorophores	46
1.3.5 Photochemical recognition of cations	46
1.3.6 Photochemical recognition of neutral guest species	48
1.3.7 Photochemical recognition of anions	50
1.4 Aims	52
Chapter 2 - Experimental	
2.0 Instrumentation and procedures	53
2.0.1 Electrochemistry	53
2.0.2 Spectroscopy	57
2.0.3 Chemicals and reagents	59
2.1 Synthesis	59
Chapter 3 - Acyclic Ruthenium(II) Bipyridyl Receptor Molecules	71
3.0 Introduction	71
3.1 Anion complexation studies	74
3.1.1 ¹ H NMR spectroscopy study	74
3.1.2 UV-vis and emission spectroscopy	75
3.1.3 Electrochemical recognition	86
3.2 Polymer modified electrode studies	92
3.2.1 Electropolymerisation	93
3.2.2 Emission spectroscopy	98
3.3 Polymer modified electrodes as anion filters	103
3.4 Conclusions	105

Chapter 4 - Macrocyclic and Calix[4]arene Ruthenium(II) Bipyridyl Receptor Molecules	107
4.0 Introduction	107
4.1 Spectroscopic anion complexation studies	109
4.1.1 ¹ H NMR spectroscopy	109
4.1.2 UV-vis and emission spectroscopy	109
4.1.3 Fluorescence emission in DMSO	113
4.2 Solvent complexation studies	117
4.2.1 Emission spectroscopy in acetonitrile on the addition of DMSO to 3	117
4.3 Electrochemical complexation study	119
4.3.1 Cyclic voltammetry	119
4.4 Conclusion	125
Chapter 5 - Diquat Based Receptors	127
5.0 Introduction	127
5.1 Metal cation complexation studies	130
5.1.1 UV-vis spectroscopy	130
5.1.2 Electrochemistry	142
5.2 Self-complexation and diquat complexation	147
5.2.1 ¹ H NMR spectroscopy	148
5.2.2 UV-vis spectroscopy	150
5.3 Anion complexation studies	151
5.3.1 UV-vis spectroscopy	152
5.3.2 ¹ H NMR spectroscopy	154
5.4 Conclusions	159
Chapter 6 - Electrochemical and spectrochemical studies of N-phenylaza-15-crown-5	160
6.0 Introduction	160
6.1 Cyclic voltammetry of N-phenylaza-15-crown-5	162
6.2 Cyclic voltammetry of N-phenylaza-15-crown-5 with addition of pyridine	169
6.3 Electrochemical recognition of metal cations	170
6.4 Electrogeneration of dimeric products and electrochemical recognition studies	179
6.5 Conclusions	183
Chapter 7 - Development of an Optical Chemical Sensor	185
7.0 Introduction	185
7.1 Instrumentation	187
7.2 Results and Discussion	190
7.3 Conclusions	195

Chapter 8 - Summary of Conclusions

8.0 Summary of Conclusions	196
References	199
Appendix 1 - Equation for the determination of stability constants from emission data	207
Appendix 2 - Equation for the determination of stability constants from absorbance data	209
Appendix 3 - List of papers published	211
Appendix 4 - Conferences attended and level of participation	212

Abstract

The aim of the project was to extend the field of molecular recognition of anions and cations of biochemical, medical, chemical and environmental importance. This was achieved by the use of a number of novel receptor molecules that are designed to bind anionic and cationic guests. The binding of the guest anions and cations was probed by various electrochemical, spectrochemical and ^1H NMR spectroscopy techniques. The receptor molecules studied included (i) ruthenium(II) trisbipyridyl complexes of acyclic, calix[4]arene and cyclic 2,2'-bipyridine ligands, (ii) the macrocycle N-phenylaza-15-crown-5, and (iii) crown ether derivatives of diquat. These receptor molecules have been shown to sense anions and cations, such as chloride, bromide, dihydrogen phosphate, sodium, lithium and magnesium. Interestingly, one of diquat crown ether derivatives has been shown to complex both anions and cations at the same time. This is an important development, as the simultaneous molecular trapping of anions and cations in such systems is seen as a possible alternative to the use of ion exchange resins.

With a view to producing spectrochemical and/or electrochemical sensor systems, work has been completed by the immobilisation of the novel receptor molecules. This has been achieved by the electropolymerisation of vinyl-substituted ruthenium(II) trisbipyridyl complexes or the immobilisation of receptor molecules in a polyvinyl chloride (PVC) matrix. The latter technique has been particularly successful and has led to the construction of a battery-powered fluorescence detector which has been used for anion sensing.

Acknowledgements

First, I would like to express my gratitude and appreciation to my supervisor, Dr. Roger Mortimer, for his invaluable advice, support, excellent guidance throughout the research and the opportunity to attend enlightening conferences. For financial support I would like to thank the Trustees of the Analytical Chemistry Trust Fund of the Royal Society of Chemistry for the award of an SAC research studentship.

I would like to extend my thanks to all my colleagues at the University of Oxford, Dr. Paul Beer, Dr. Fredrich Szemes, Dr. Dusan Heseck, Dr. Andrew Graydon and also Dr. Oldrich Kocian at the University of Birmingham. I would also like to thank the technical staff from the Chemistry Department in particular Alan Stevens, John Spray, Dave Wilson, Simon Riggott, Bev Cooper and Elaine Till for technical assistance and John Kershaw for the ^1H NMR spectroscopy data.

I wish to express my sincere gratitude to a good friend and colleague Prof. Nelson Stradiotto (University of São Paulo), whose assistance at the start of this project was invaluable, I would like to extend this gratitude to Dr. Alan Davies, Dr. Dave Worrall and Dr. Stephen Summerfield for many fruitful discussions.

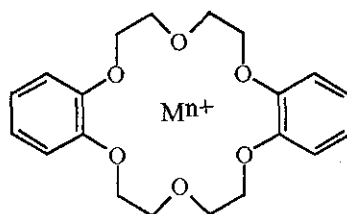
I would like to thank all my friends throughout my time here in Loughborough, in particular, the 'polymer lads', Deb Hall, Mike Simcox, Kirk Lewis, Stephen Webster, Norman Wood, Hilary Evans and Andrew Kane.

Finally, I would like to thank Alison, my parents and my sister Sarah who have continually been a source of ideas, support and encouragement.

Chapter 1 - Introduction

1.0 Introduction

For more than 150 years, organic chemists were predominantly concerned with the nature of the covalent bond in organic molecules. This was, however, changed by the original work carried out by Pedersen, who by chance discovered dibenzo-18-crown-6 **1** in 1967.¹ Subsequent discoveries^{1,2} that **1** and other macrocyclic polyethers, the crown ethers, selectively complex biologically relevant alkali and alkaline earth metal cations led to a rapid increase in the development of the field of synthetic host-guest chemistry. The original and inspiring research performed by Lehn^{3,4} on the novel bicyclic cryptands and Cram⁵ on the chiral crown ethers and the rigid spherands culminated in the award of the Nobel prize for Chemistry to Pedersen, Lehn, and Cram in 1987. The results of the aforementioned researchers and others will be discussed in further detail.



M^{n+} = metal cation

1

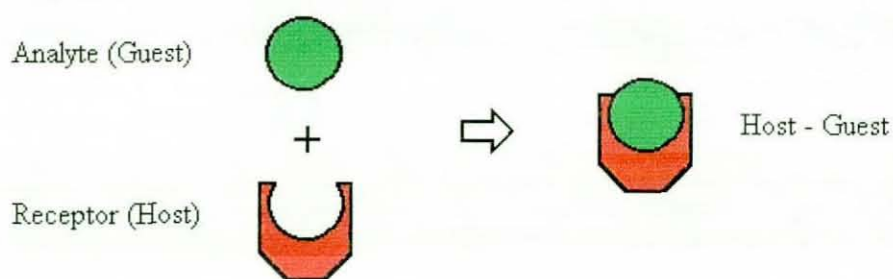
Pedersen's discoveries led to the formation of a new field of chemistry, which for the first time inspired organic chemists to venture into the study of non-covalent bonding of molecules.

This area has been entitled **Supramolecular Chemistry**^{4,6} which can be defined as the study of the results of intermolecular bonding of two or more molecules. The resulting new complex or adduct, can simply be defined as a supramolecule; a “molecule” of molecules. The source of intermolecular bonding is electrostatic in nature, and includes hydrogen-bonding, ion-pairing, π -acid to π -base interactions, metal-to-ligand binding and van der Waals' forces. By the correct manipulation of the energetic and stereochemical features of such non covalent bonds during the design of artificial, abiotic receptor molecules, it is possible to design receptors with high efficiency and selectivity.

In the supramolecular approach, **selectivity** can be designed into the **host** to give **molecular recognition**. This is achieved by altering the binding between the host and the guest by utilising the following principles.

- a) **Steric factors**:- matching the dimensions of the cavity of the host molecule to that of the target guest
- b) **Binding sites**:- incorporating binding sites into the cavity of the host molecule such that they are complementary in number, position in space and features of the target guest

The design of a host molecule to recognise a specific guest will result in the required selectivity, a sort of lock and key approach (**Scheme 1**).



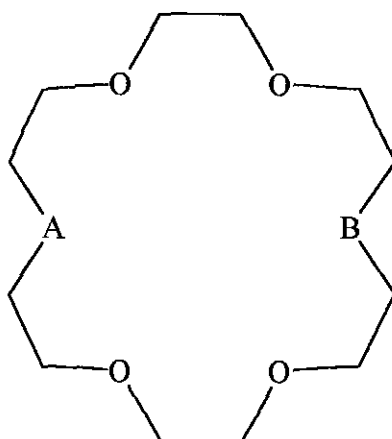
Scheme 1 Ideal molecular recognition is where for a given analyte, a receptor molecule is designed which possesses geometrical and bonding features which give a specific interaction.

A more specific terminology for this new area is **Host-Guest Chemistry**.⁷ The host is generally a large organic molecule whereas the **guest** is generally a small cation, anion, neutral molecule or atom. Nature has been generous and has provided a large abundance of guest molecules to choose from, whereas the host receptors generally have to be designed and synthesised. It is important when a host receptor is designed that it is either specific to a particular guest molecule or more commonly specific to a limited number. Therefore it is important to consider the characteristics of the target guest molecule in the receptor design process. With some receptor molecules, for example crown ethers, the specificity to alkali and alkaline earth metal cations is due to the size of the binding site. This results in selectivity due to size exclusion.

It is also worth noting that it is the donor atom at the binding site which dictates the nature of the host-guest interaction. Frensdorff⁸ found that changing the co-ordination atom can lead to quite subtle changes in properties. The replacement of oxygen as the donor atom by nitrogen or sulphur in an 18-crown-6 ligand leads to a decrease in stability and selectivity for the alkali and alkaline earth metal cations. This is principally

due to binding in the alkali and alkaline earth metal cations taking place by electrostatic interaction. The electronegative charge on the heteroatom drops $O > NR > NH > S$, which leads to a reduction in electrostatic interaction. However, in the case of Ag^+ , covalent bonding is involved. **Table 1** shows that complexation properties can be altered by changing the nature of the binding site.

Table 1 Effect of N and S substitution on the stabilities of 18-crown-6 complexes



Binding-site atom		log Stability constant (K_s)	
A	B	K^+ (MeOH, 25°C)	Ag^+ (H ₂ O, 25°C)
O	O	6.10	1.60
NH	O	3.90	3.3
NH	NH	2.04	7.8
S	S	1.15	4.34

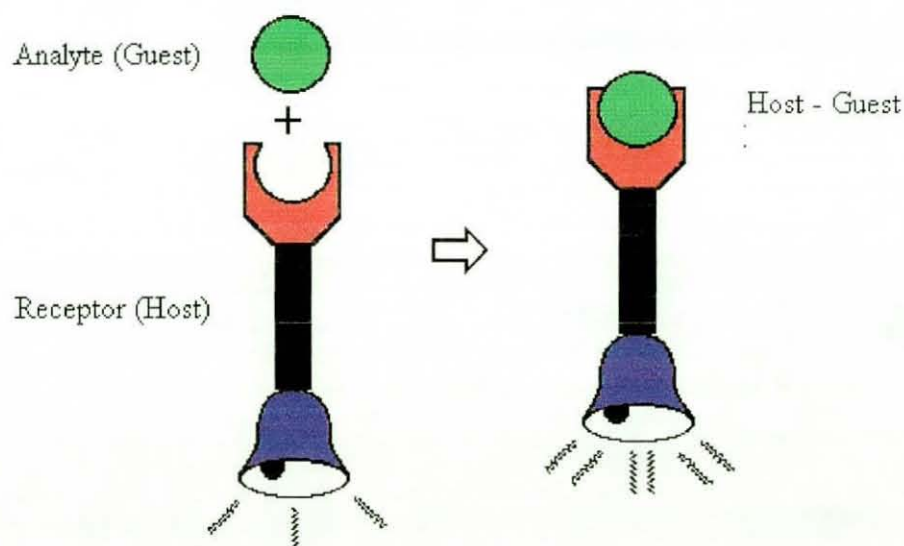
The thermodynamic stability constant for a 1:1 complex, K_s , is defined by

$$K_s = [M^{+}Cr] / [M^{+}][Cr]$$

where $[M\text{Cr}^+]$, $[M^+]$ and $[\text{Cr}]$ are the molar concentrations of complexed cation, uncomplexed cation and uncomplexed polyether respectively.

Host molecules as sensors

Host molecules can act as a chemical sensor if they undergo some detectable physical change on the aforementioned recognition and binding of a guest. This can be achieved by the incorporation of a signalling unit in combination with a receptor molecule (**Scheme 2**). A change in physical properties on complexation can be detected by a number of different approaches. Possible methods of detection include electrochemistry if the molecule contains a redox active function,⁹ UV-visible spectroscopy of a chromophore¹⁰ or more recently fluorescence emission on the incorporation of a fluorophore¹¹ such that perturbations in the excited-state energies can be measured.



Scheme 2 The combination of a receptor and a signalling unit produces a sensing receptor

The remainder of the introduction is mainly dedicated to the developments made in the design of abiotic receptor molecules for cations, anions and neutral species, which leads on to a detailed discussion of the utilisation of the receptor molecules in the electrochemical and spectrochemical detection of guest species. It is however important to remember that Nature was the original and exquisite designer of receptor molecules. Therefore a small section is devoted to biotic receptors and the large role they play in inspiring the design of receptors.

1.1 Biotic (natural) and abiotic (synthetic) receptors

Biotic receptors - Nature the original designer

Nature is full of biotic receptors which are elegantly demonstrated by enzyme-substrate interactions, biosynthesis of proteins, and antigen-antibody reactions. Many cellular processes require, as an initial event, the specific recognition and binding of biologically active molecules. The receptor molecules responsible for recognition processes are generally proteins. These protein receptors control biological systems by binding regulatory molecules. This generally induces a conformation change which can lead to activation or inhibition of an enzymic activity, with resulting changes in cellular metabolism. Numerous other highly specific biological processes take place due to intermolecular interactions. These include the formation of protein complexes, the intercalation of complexes of nucleic acids, the decoding of the genetic code and neurotransmission. The discovery of natural macrocyclic receptors such as the cyclodextrins and valinomycin, played a crucial role in the birth of the chemistry of molecular recognition.

Cyclodextrins

α -, β -, and γ -Cyclodextrins are cyclic oligosaccharides consisting of six, seven, or eight glucose units, which were first isolated in 1891 by Villiers as degradation products of starch.¹² Cyclodextrins are rigid in structure and bucket-shaped, complexation takes place by the complexed molecule filling the cavity of the cyclodextrin. The hydrophobic interior and hydrophilic exterior provide a place for organic molecules such as benzene (Fig. 1) to “keep dry” in aqueous solution.

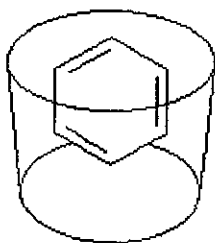


Fig. 1 Representation of the Cyclodextrin-benzene complex

Protoporphyrin IX - receptor for neutral molecules

Probably one of the most important biological receptors is the life giving porphyrin protoporphyrin IX and its iron (II) complex haem (Fig. 2).¹³ The iron complex of protoporphyrin IX provides a site of reversible oxygen binding in myoglobin and haemoglobin which is found in vertebrates and many invertebrates. Myoglobin and haemoglobin store oxygen until it is required for biological processes such as the oxidative phosphorylation of ADP in the mitochondria, which are present in all living cells. A number of research groups are attempting to mimic this porphyrin with the aim of designing a sensor for the detection of carbon monoxide. Carbon monoxide

poisoning takes place because it is preferentially bound to the binding site preventing the take up of oxygen.

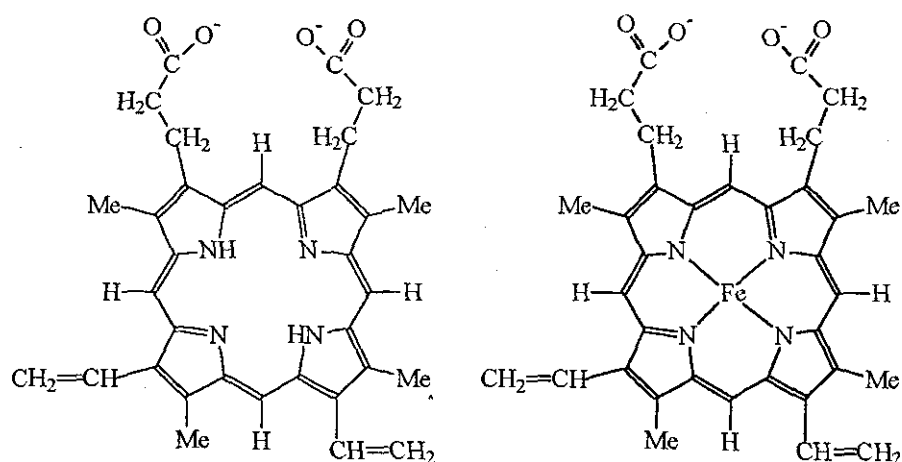


Fig. 2 Protoporphyrin IX and its iron (II) complex haem.

Valinomycin

The natural receptor valinomycin (**Fig. 3**) was first isolated from *Streptomyces fulvissimus* by Brockmann and Schmidt-Kastner in 1955.¹⁴

Valinomycin is an ionophoric antibiotic, which means that it can facilitate the passage of inorganic cations across membranes.¹⁵ This macrocycle forms a well-defined complex with potassium ions and the dimensions are such that the potassium ion is exactly accommodated. Although valinomycin will also form a complex with sodium the smaller sodium atom fits much less exactly into the structure and this complex has a stability constant one thousand times smaller than that of the potassium complex.¹⁵ The binding of the potassium ion in the structure of valinomycin increases the lipophilicity of the antibiotic and thereby promotes its diffusion into the hydrophobic regions of the membrane. Valinomycin's antibiotic nature is due to its ability to specifically drain the

cell of potassium. The growth of the cell ceases due to the requirement of potassium in cellular metabolism. Valinomycin was discovered because of its antibacterial properties, but it is not used in medicine due to its lack of specificity.

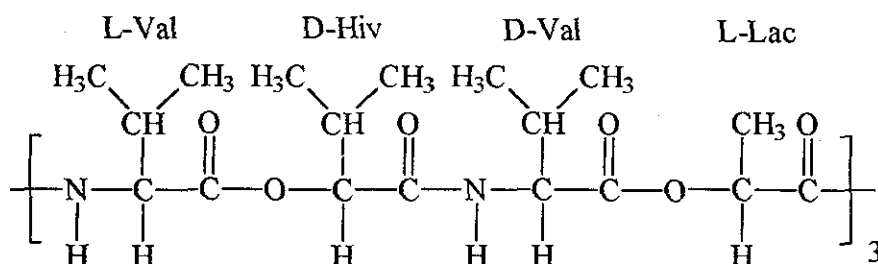


Fig. 3 Structure of valinomycin is composed of three repetition units of Val (Valine), Lac (Lactic acid) and Hiv (2-hydroxyisovaleric acid). This antibiotic affects the permeability of membranes to potassium ions.

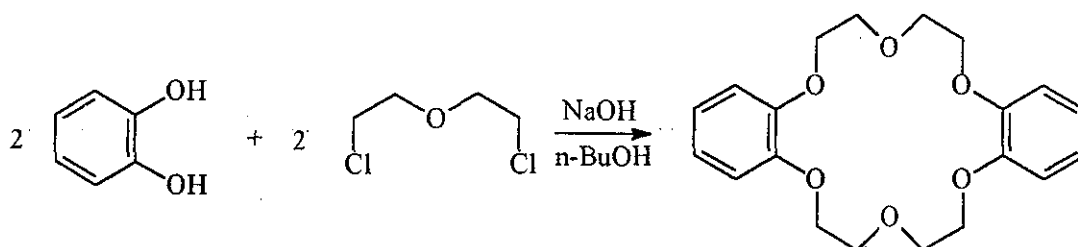
1.2 Abiotic receptors

1.2.1 Receptors for cationic guest species

Crown ethers

Crown ethers were discovered quite by chance whilst Pedersen was investigating the catalytic activity of vanadium in oxidation and polymerisation reactions for Dupont chemicals.^{1, 2} Whilst researching the aforementioned area, Pedersen produced a small quantity of an unknown compound. Fortunately due to Pedersen's curiosity, a series of experiments were performed on the unknown compound with the discovery that the compound had a low solubility in protic solvents.

The unknown compound was subsequently shown to be 2,3,11,12-dibenzo-1,4,7,10,13,16-hexaoxacyclooctadeca-2,11-diene, more commonly known as dibenzo-18-crown-6, which was formed by the reaction of bis(2-chloroethyl)ether with unprotected catechol (**Scheme 3**).



Scheme 3 Formation of dibenzo-18-crown-6

A curious discovery was made when the low solubility of the crown ether in protic solvents was found to be overcome by the addition of sodium ions. Pedersen postulated that the sodium was bound into the cavity and held there by the electrostatic attraction between the positive charge of the sodium and the negative dipolar charge on the six oxygen atoms. An even larger effect was seen on the addition of potassium iodate in methanol at 30°C. This gave a 38 fold increase in the solubility of dibenzo-18-crown-6.

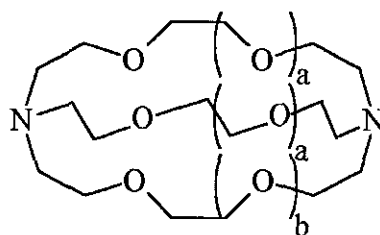
A number of crown ethers were then synthesised with cavities containing four to ten oxygen atoms. On changing the number of oxygen atoms in the ring it is possible to demonstrate selectivity of a certain crown ether for a particular alkali or alkali earth metal cation as shown in **Table 2**.¹⁶ Selectivity was found to be due to the cavity size and the metal cation diameter being complementary, resulting in thermodynamically stable complexes.

Table 2 Cation binding strengths for various crown ethers

Stability Constants, log K _s			
Ligand	Na ⁺	K ⁺	Ca ²⁺
12-crown-4	1.7	1.3	—
15-crown-5	3.24	3.43	2.36
18-crown-6	4.35	6.08	3.90

Lehn's novel bicyclic cryptands

Cryptands³ were developed and originally produced by Lehn, their development was seen as an innovating progression from Pedersen's crown ethers. They contain a three-dimensional cavity which is varied in size by altering the lengths of the bridges such as in **2**. Unlike the crown ethers, they offer a three-dimensional structure which results in greater selectivity and provides stronger complexation of the cation by entirely encasing the cation guest.



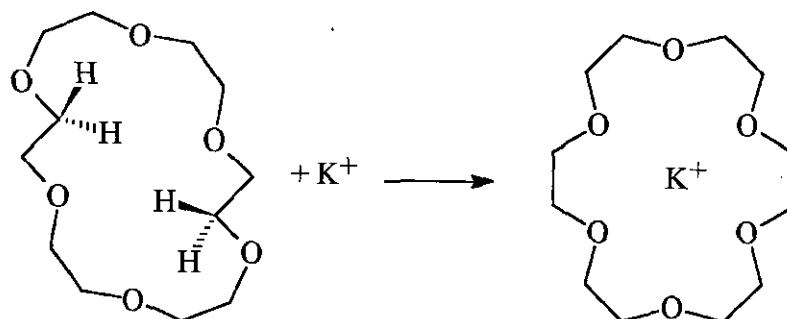
a=1, b=1 [2,2,2]Cryptand
a=1, b=0 [2,1,1]Cryptand
a=2, b=2 [3,3,3]Cryptand

For example, the potassium ion-[2,2,2] cryptate is 10^4 times more stable than the potassium ion complex of 18-crown-6, the crown ether of comparable size.¹⁷ This increased binding stability lies in the cryptate effect and is of an enthalphic origin.

The name cryptand was derived from the Greek word cryptos meaning cave.

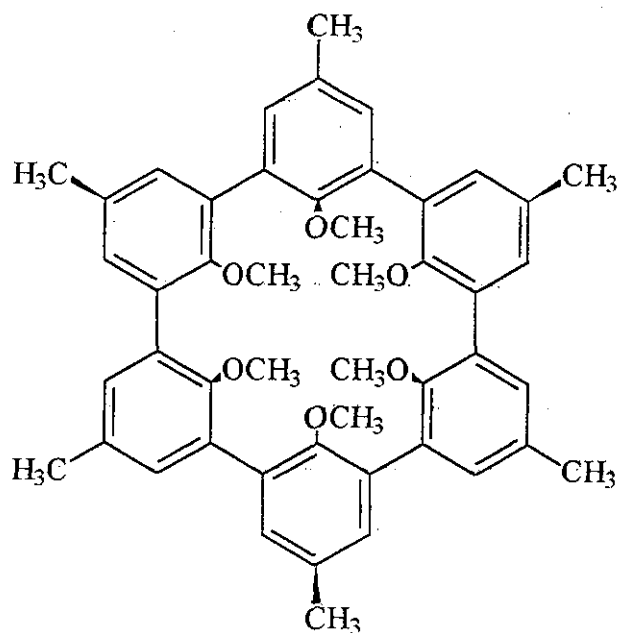
Cram's chiral crown ethers and rigid spherands

Analysis of the crystal structures of the free crown ethers and cryptands gave rise to spherands such as **3**. Structural analysis of these crown ethers and cryptands showed that on complexation there is a reorganisation of these ligand binding sites. Thermodynamically any conformational reorganisation disfavour the formation of the complex to some extent. Crystal structure analysis of Pedersen's 18-crown-6¹⁸ and Lehn's [2,2,2]cryptand^{19, 20} shows that in their uncomplexed states, they contain neither cavities nor convergently arranged binding sites. This is typified by comparing the crystal structure of the host (**Scheme 4**) with that of its K^+ complex,⁵ which indicates that the complexing act must be accompanied by host reorganisation and desolvation.



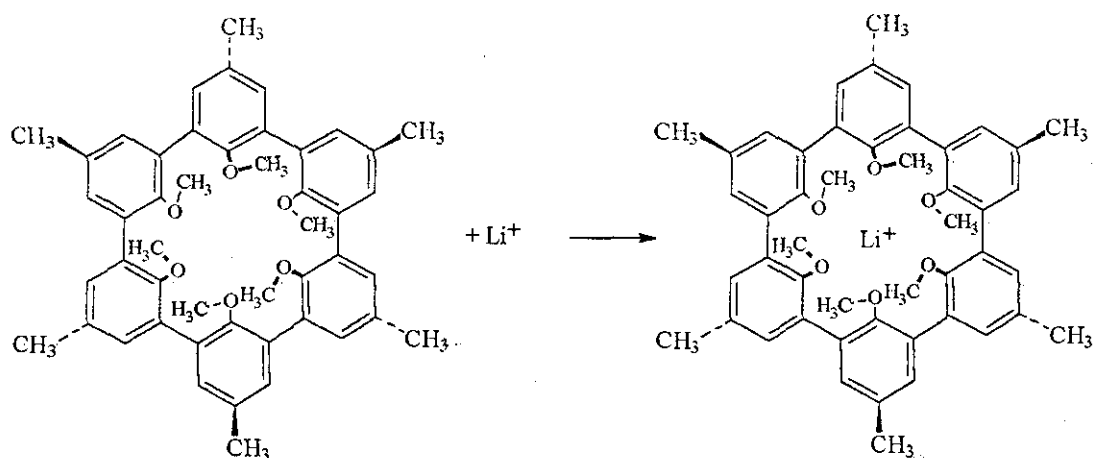
Scheme 4 Conformational rearrangement of 18-crown-6 on binding of potassium cation

Spherands were synthesised by Cram, with the intention of preorganising the binding sites of the receptor, thus removing the requirement for any conformational modification.²¹



3

The crystal structure of the spherand in (Scheme 5) and its Li^+ complex were compared to determine conformational changes on complexation.²² As proposed there were minimal conformational change on complexation.



Scheme 5 Crystal structure analysis of a spherand on complexation with Li^+

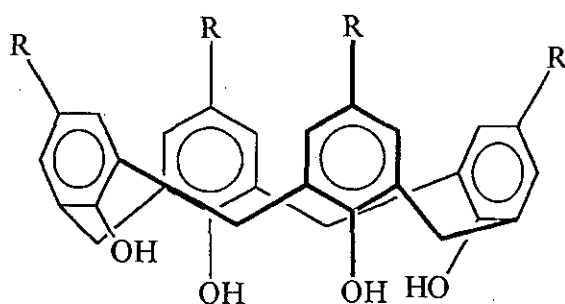
In fact these molecules show the highest stability constants known for complexes of many alkali and alkaline earth metal cations, and also great selectivity. For example, the spherand in (Scheme 5) has a selectivity factor (ratio of stability constants) of 600 for Li^+ over Na^+ , and will not bind K^+ at all as the cavity is too small.

1.2.2 Receptors for neutral guest species

Calixarenes - the molecular basket

During the past decade a group of phenolic macrocycles known as the calixarenes have had growing significance in supramolecular chemistry.^{23, 24} However the origins of the calixarenes can be traced back to the 1870's with the discovery of phenol-formaldehyde resins by von Baeyers. The polycondensation of phenol and formaldehyde in acid or base proceeds through the ortho and para positions, leading ultimately to the highly cross linked matrix characteristic of Bakelite. It was discovered that if the para position

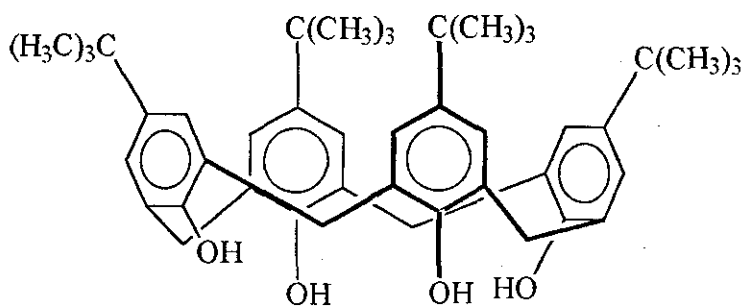
of the phenol was blocked the condensation reaction between formaldehyde and para-substituted phenol led to the isolation of macrocyclic oligomers. A simple example of such a calixarene is the cyclic tetrameric compound **4**. The name calixarene is derived from the Greek calix (a chalice or cup), this is due to the molecular model having a shape similar in appearance to a Greek crater vase.



R = same or different

4

The most useful property of the calixarenes comes from their ability to bind neutral and ionic guests within supramolecular arrays. Simple phenol-derived calixarenes such as p-tert-butylcalix[4]arene **5** form complexes with neutral organic molecules including chloroform, benzene, xylene and anisole.^{25, 26}

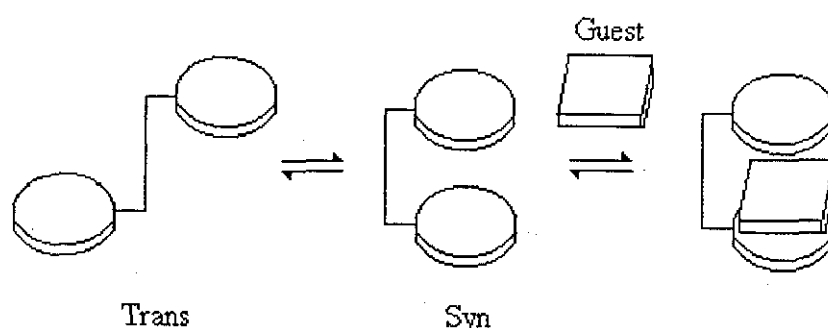


5

A number of research groups are working on the development of calixarene based sensors. Complexation by the calixarene is probed using electrochemical and optical techniques.²⁷

Molecular tweezers for neutral molecules

A number of workers have been synthesising and investigating the association abilities of receptors for neutral aromatic molecules. The neutral aromatic molecule is bound due to the formation of a π -system stacking sandwich complex. The receptor molecules are generally referred to as "molecular tweezers" (Scheme 6).²⁸

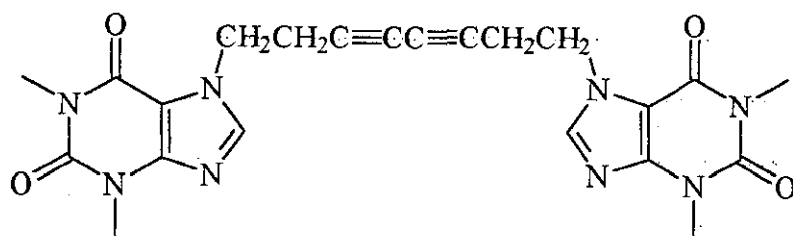


Scheme 6 Molecular Tweezers as receptor for neutral guest - complexation can only take place in the syn conformation

Ideally these receptor molecules must satisfy three criteria to enhance complexation of aromatic molecules in aqueous solution:-

1. rigid spacer units should be present to prevent self-association of the receptor units
2. the distance between the π -system in the receptor molecule must be sufficient for the proper insertion of a π -system guest between the rings.
3. a rigid syn conformation

Chen and Whitlock have synthesised several bifunctional derivatives of caffeine such as **6** which complex aromatic molecules.²⁹ The molecules act as molecular tweezers and lead to the formation of a π -system hydrophobic complex with molecules such as 2,6-dihydroxybenzoate (DHBA) or 1,3-dihydroxy-2-naphthoate (DHNA). Molecules were designed such that the rigid diyne unit prevents self association of the two caffeine moieties. It is also crucial that the distance between the aromatic ring is sufficient to allow the insertion of a guest molecule. However a rigid syn conformation is not obtained. A rigid syn conformation enhances the complexation ability of the tweezer molecule.



6

Rebek and co-workers have developed a number of rigid syn conformation receptors based on an aromatic backbone.^{12, 30} In the case of the acridine spacer group (**Fig. 4**) a large flat π -bonded surface is presented. This allows the aromatic groups of the host and the guest to undergo π -stacking. Rotation around the $C_{\text{aryl}}-N_{\text{imide}}$ bond is impossible due to the methyl group on the acridine. This results in the molecule being locked in the syn conformation which leads to great structural rigidity and binding enhancement. The binding of the lactams involves hydrogen bonding where the amide components must be either amides or lactams.

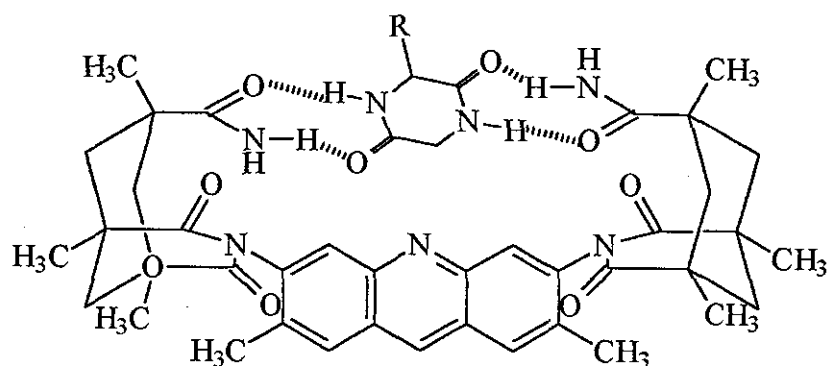


Fig. 4 Hydrogen bonding in molecular tweezers

1.2.3 Receptors for anionic guest species

This thesis is mainly concerned with the study of the complexation of anionic guest species, therefore anion receptors will be treated in detail in this introduction. The complexation of anion guest species is a relatively unexplored area, when compared against studies on the complexation of metal and molecular cations, and increasing research into hosts for neutral molecules.

Anions are of critical importance in the fields of biochemical, medical and environmental sciences. It is therefore very surprising that the molecular recognition of anions have received comparatively little attention during the lifetime of supramolecular chemistry.³¹

Nature however has made great use of anions, which are necessary partners to positive centres (ammonium, guanidinium, metal cations etc.) where their functions are of equal importance to those of cations. In fact the majority of natural enzymes (ca 70%) are known to bind anionic guest species.

Physical properties offer an explanation into why so little work has been attempted in the field of anion complexation:-

1. Anion sizes are relatively large with respect to cations (Tables 3 and 4 respectively).³² The smallest anion is F^- (1.36 Å) which is about the same size as K^+ (1.38 Å), therefore larger host molecules must be developed for anions than cations.

Table 3 Anion radius Å³²

F^-	1.36	OH^-	1.40	NO_2^-	1.55	CO_3^{2-}	1.85
Cl^-	1.81	CN^-	1.82	NO_3^-	1.89	SO_4^{2-}	2.30
Br^-	1.95	IO_3^-	1.82	MnO_4^-	2.40	PO_4^{3-}	2.38
I^-	2.16						

Table 4 Cation radius Å³²

Li^+	0.73
Na^+	1.02
K^+	1.38
Cs^+	1.67

2. The Periodic Table disfavors the complexation of anions with the lack of suitable receptors. Cations on the other hand have a variety of donor atoms such as O, S, N and P which are available for complexation. However, receptors can be provided by

using neutral binding sites such as -OH or -SH, or positively charged ligands containing, for example, ammonium or guanidinium moieties.^{33, 34}

3. The molecular shape of cations are generally spherical, whereas anions may occur in a wide variety of geometries,³⁵ e.g. $\text{Fe}(\text{CN})_6^{4-}$ is octahedral, PO_4^{3-} is tetrahedral and N_3^- is linear (Table 5).

Table 5 The geometries of some anions³⁵

Spherical	Planar	Linear	Tetrahedral	Octahedral
F^-	NO_3^-	N_3^-	PO_4^{3-}	$\text{Fe}(\text{CN})_6^{4-}$
Cl^-	CO_3^{2-}	CN^-	SO_4^{2-}	$\text{Co}(\text{CN})_6^{3-}$
Br^-	RCO_2^-	SCN^-	ClO_4^-	
I^-			MnO_4^-	

4. The solvation energies of anions are much larger than that for cations of similar size.³⁶ For example, K^+ has a solvation energy in water at 25°C of 337.2 kJ mol⁻¹ whereas for F^- it is 434.3 kJ mol⁻¹. Complexation is in fact in competition with solvation. The increase in solvation energies for anions (Table 6) makes them harder to complex than cations due to the extra energy required on removal of a solvation shell.

Table 6 The solvation energies (ΔG) of some ions (in water at 25°C/kJ mol⁻¹)³⁶

Cation	$r/\text{\AA}$	ΔG
Li ⁺	0.73	510.9
Na ⁺	1.02	410.9
K ⁺	1.38	337.2
Rb ⁺	1.67	315.9
Cs ⁺	1.86	287.0

Anions	$r/\text{\AA}$	ΔG
F ⁻	1.36	434.3
Cl ⁻	1.81	317.1
Br ⁻	1.95	303.3
I ⁻	2.16	256.9

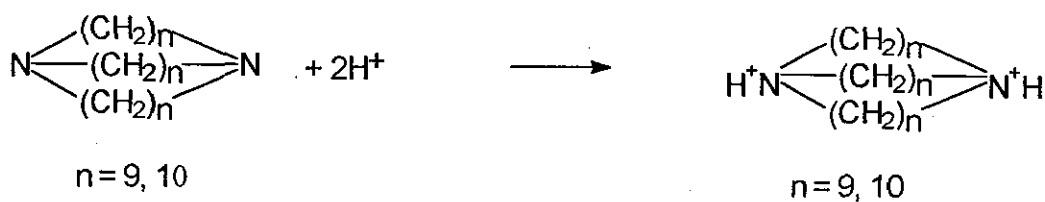
5. A vast proportion of anion molecules exist over a limited pH range e.g. carboxylates exist only at pH's above 5-6.³⁷ It is therefore critical that the host molecule has the ability to complex in the pH region where the guest is anionic.

The binding sites of host molecules generally contain neutral receptor sites, e.g. -OH, or -SH, or positively charged ligands containing, for example, ammonium or guanidinium moieties. This section will report on classes of compounds including protonated polyamines, guanidinium cations and Lewis acid-containing ligands. The anion complexed by the host molecules will also be detailed.

Receptors with ammonium centres

Macropolycycles

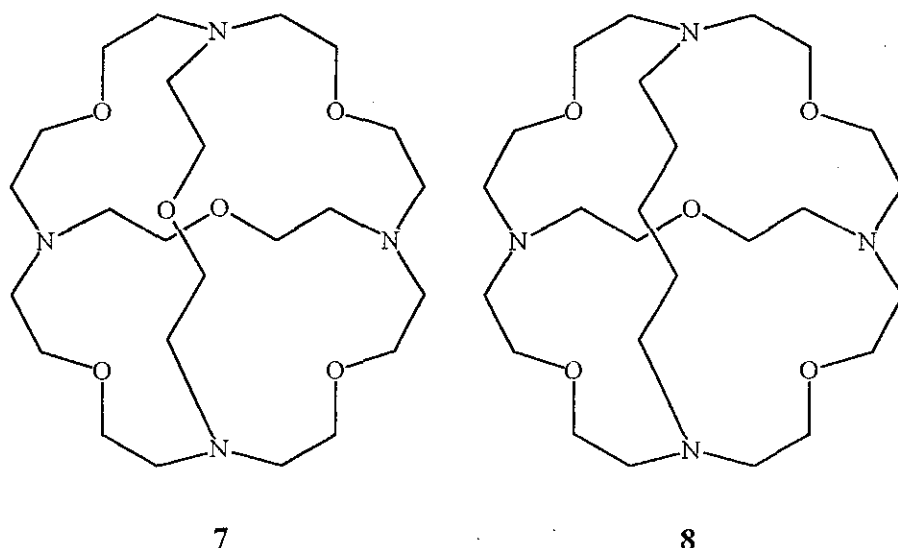
Synthetic anion complexing agents (**Scheme 7**) were first reported by Park and Simmons in 1968.³⁸



Scheme 7 Macropolycycles which complex anions

Protonation of the above molecule leads to the diprotonated form shown, with both hydrogens facing into the cavity. Such diazamacrobicycles bind halide ions (Cl^- , Br^- , and I^-) producing stable complexes. As with the crown ethers, the selectivity towards a particular halide depends on the cavity size. This results in spherical anion recognition. Stability of the complex results from the interaction of the positive charge as well as hydrogen-bonding within the cavity giving $\text{N}^+\text{-H}\cdots\text{Cl}^-\cdots\text{H-N}^+$.

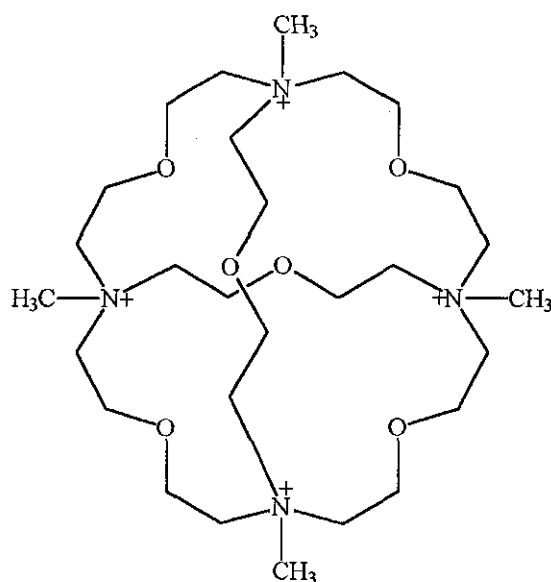
Macropolycycles were further developed by Lehn, who synthesised the macrotricyclic anion cryptates **7** and **8**.³⁹ These receptors have quaternary binding sites and a cage like structure which induces high selectivity.



In the tetraprotonated form they are highly stable and provide selective complexes for the spherical halide anions.³⁹ For example with $8-4H^+$ there is high selectivity for Cl^- over Br^- (>1000) as calculated from stability constants. Selectivity is dependent on the cavity size.

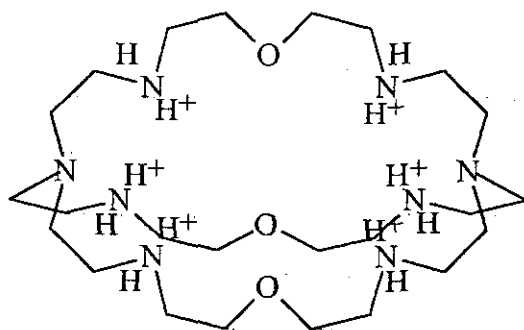
The X-ray crystal structure shows that the Cl^- is held within a tetrahedral array of hydrogen bonds $N^+-H\cdots Cl^-$.⁴⁰

Receptor **7** was resynthesised by Schmidtchen to incorporate methyl groups at the nitrogen in place of the protons to give **9**.⁴¹ By carrying out this modification, halide ion complexes of this receptor can only be formed by electrostatic interactions other than hydrogen-bonding. Subsequently, stability constants were obtained for this complex which were shown to be significantly lower than the equivalent tetraprotonated receptor, thus indicating that quaternary ammonium salts are less effective at binding anions than polyamines due to the lack of hydrogen bonding.⁴¹ However, Br^- was shown to be more strongly bound than Cl^- due to the larger intramolecular host cavity present in the receptor.



9

So far only receptors such as 7-9 have been considered that bind spherical halide anions. This is because until about 15 years ago very few synthetic receptors were able to accommodate other anionic geometries, due to the complex synthesis procedures for receptors which have non-spherical cavities. In 1978 the macrobicyclic receptor 10 was designed for the recognition of linear triatomic species.⁴² As with the previous examples, anion complexation takes place due to hydrogen bonding when the receptor is present as the hexaprotonated form. Protonation occurs at the six secondary nitrogen sites in the bridges. The ellipsoidal cavity provides a binding site for linear anions such as azide (N_3^-) ion. The receptor also has an important feature which is the stabilisation of normally unstable species such as F_2H^- , Cl_2H^- , F_3^- , Cl_2F^- , etc.



10

In 1991 this idea was further developed to produce a receptor molecule, which in the hexaprotonated form binds dianionic guests.⁴³ In fact it produces a very stable complex with the terephthalate anion (**Fig. 5**), indicating that there is a close correlation between the cavity size and the terephthalate anion size. Effective complexation is due to the combined electrostatic and hydrophobic effects between the host and the guest

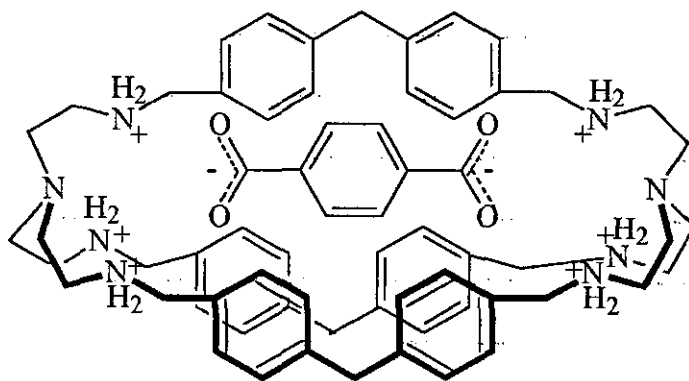
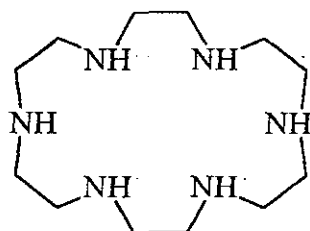


Fig. 5 Complexation of a terephthalate anion

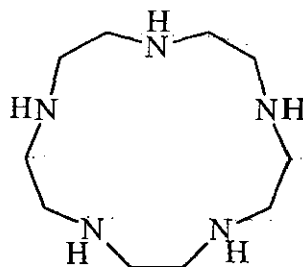
Macrocycles for anions

Simple polyazamacrocycles, such as **11** and **12**, in their fully or partially protonated forms will bind anions via electrostatic and hydrogen-bonding interactions. However,

with receptor **13** numerous anionic species, for example NO_3^- , Cl^- , ClO_4^- , Br^- , CF_3CO_2^- and IO_3^- can be bound due to the larger cavity size.⁴⁴⁻⁴⁷

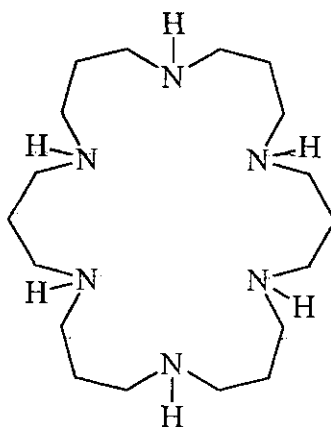


11



12

One disadvantage found with hexacyclens such as **11**, **12** and other polyazamacrocycles where the nitrogen atoms are separated by ethylene groups, is that full protonation occurs only at low pHs, where many anions are also protonated. By reducing electrostatic repulsion within the receptor, full protonation can occur at higher pHs. This can be achieved by separating each nitrogen by three or four methylene groups such as in **13**.⁴⁸



13

Receptor 13 is fully protonated at neutral pH's and forms strong complexes with polyanions like adenosine monophosphate (AMP), adenosine diphosphate (ADP), adenosine triphosphate (ATP), sulphate and oxalate.

The discovery of the macrocyclic polyamine (Fig. 6) yielded a supramolecular catalyst capable of proto-ATPase, protokinase, and protophosphatase activity.⁴⁹

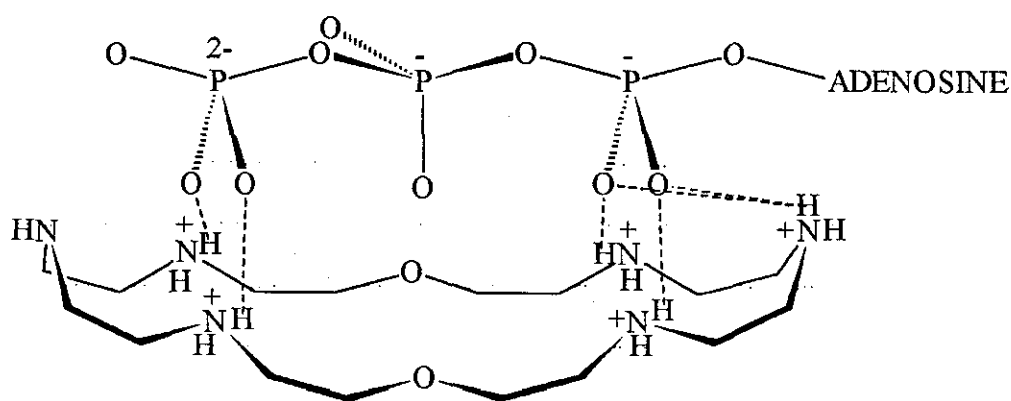


Fig. 6 Complexation of ATP by a macrocyclic polyamine

In the protonated form the anion receptor binds nucleotides in the order ATP>ADP>AMP. It acts as a catalyst converting ATP first, rapidly, into a ADP and then, more slowly, into AMP and inorganic phosphate in a pH-independent process between $3 < \text{pH} < 9$. The catalytic mechanism is not clear.

Second Sphere Co-ordination

Large polyammonium receptors molecules have been synthesised to complex $[\text{Co}(\text{CN})_6]^{3-}$, $[\text{Fe}(\text{CN})_6]^{3-}$ and $[\text{Ru}(\text{CN})_6]^{4-}$ anions via second sphere complexes.⁴⁸⁻⁵³ In



The guanidinium group occurs in nature as an anion binding site due to a number of interesting features:-

2. the guanidinium cation orientation easily forms a characteristic pair of well-organised, strong hydrogen bonds with a guest which has a complementary structure,⁵⁵ such as carboxylates and phosphates. X-ray crystallography has shown that the guanidinium salts, have the general structure shown in **Fig. 7**.

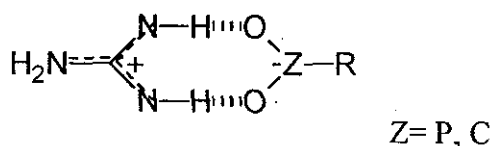
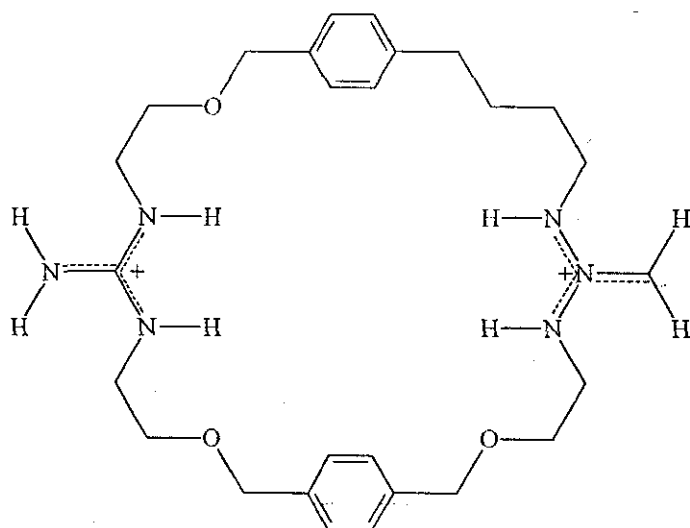


Fig. 7 Binding of guanidinium by hydrogen bonding to an oxy-anionic species

3. Guanidinium ion groups in arginyl residues of proteins are largely involved in maintaining the conformation of proteins. Also, due to the pattern of its hydrogen bonding it plays a critical role in the recognition of anions by a number of enzymes, receptor sites and antibodies.^{56, 57}

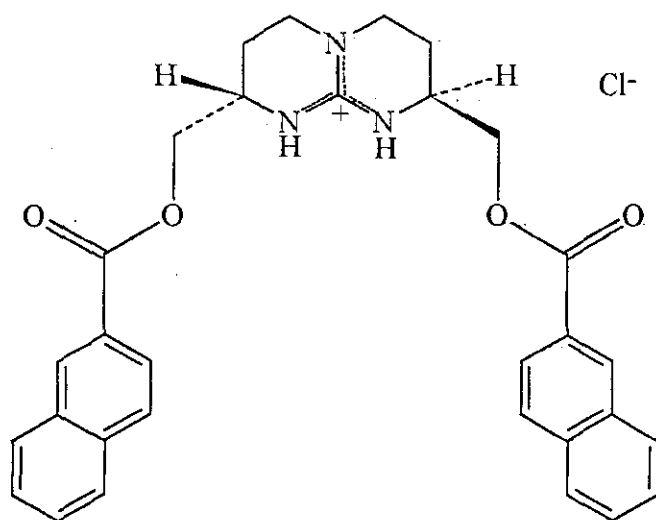
Lehn and co-workers in 1978 developed a method for the introduction of guanidinium groups into macrocyclic molecules.⁵⁸ Macrocyclic structures possess the architectural flexibility to allow the designed arrangement of anion binding sites. The macrocycle **15** formed a 1:1 complex with PO_3^- , and gave a stability constant of $\log K_s = 2.2$ in water.



15

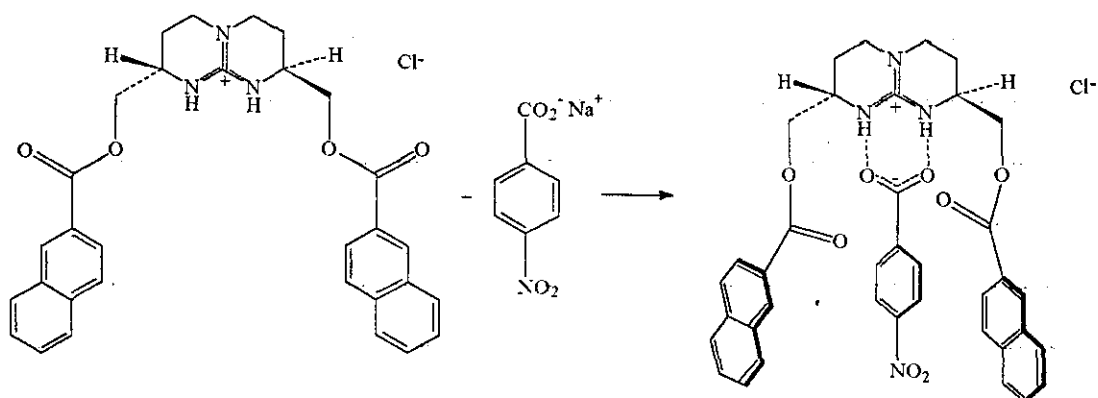
This complex shows a weak chelate and macrocyclic effect when compared against the strength of complexes formed by acyclic analogues. With this in mind, work has mainly been focused on acyclic polyguanidinium salts which are easier to prepare than macrocycles.

Receptors have been developed to include the rigid chiral guanidinium subunit shown below.⁵⁹ The guanidinium forms strong, well-organised zwitterionic hydrogen bonds ($\text{N-H}^+ \cdots \text{O}^-$) with carboxylates or phosphates. Complex **16** incorporates naphthoyl groups which are able to undergo π - π stacking with aromatic groups of the guest species, giving a complex with a well-defined geometry and double recognition of the guest. The chirality of **16** also forces the substrate to bind in a dissymmetric environment, allowing the enantioselective recognition of chiral carboxylic acids.



16

One interesting property is that it can quantitatively extract sodium p-nitrobenzoate from water into a chloroform solution of **16** (Scheme 8).



Scheme 8 Mechanism for the extraction of sodium p-nitrobenzoate in water to chloroform

Receptors containing Lewis acid binding sites

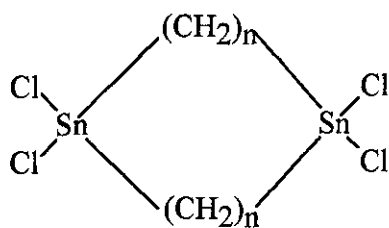
A number of groups have been working on the incorporation of Lewis acidic sites into macrocyclic and acyclic ligands. These include Lewis acidic binding sites such as tin,

boron, silicon and the uranyl cation which are incorporated into receptors for binding anions. The Lewis acidic centre acts as an acceptor site for the electron density. Presently, only a few molecules have been developed.

Lewis acid binding sites

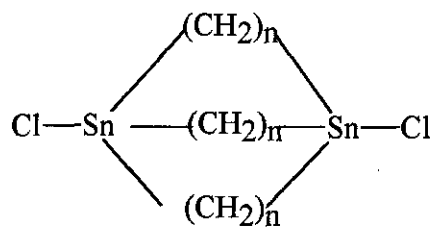
Tin

Host molecules with tin atoms acting as Lewis acid sites in macrocycles such as **17**, were some of the earliest hosts to be prepared for anions.^{60, 61} Low selectivity was observed for chloride binding by these macrocyclic hosts containing two binding sites. Newcomb and co-workers found that the incorporation of structural rigidity into the Lewis acidic macrocyclic hosts such as **18** led to greater anion selectivity.^{60, 61}



$n=8, 10, 12$

17



$n=6, 7, 8, 10, 12$

18

Rigidity rapidly increased since the Lewis acidic tin complex contains one electron withdrawing atom. This group will complex donors in a trigonal-bipyramidal structure, with the donor and withdrawing groups in the axial positions.

Boron

Reetz et al. reported⁶² a new class of anion-selective receptors, **Fig. 8**, which in addition to a conventional crown ether moiety for complexation of cations contains a Lewis acidic boron centre for complexation of anions. The intermolecular inclusion of K^+ and F^- was established using X-ray crystallography, ^{11}B and ^{13}C NMR spectroscopy. X-ray crystallography showed that the K^+ cation was complexed by five oxygens in the crown and also by one of the boron oxygens, whereas the fluoride anion is only complexed by the boron atom.

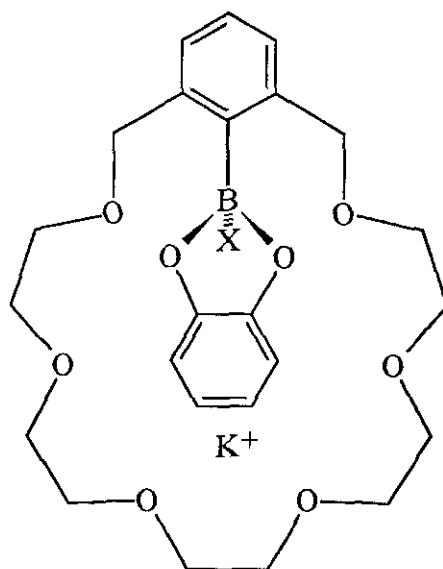


Fig. 8 Complexation of K^+ and a halide, X^- (F^-) by a boron-containing complex.

Silicon

The first crown silane was reported by Jung and Xia in 1988.⁶³ It is thought that the halide anion is complexed by the three silicon atoms, but the true nature is not known.

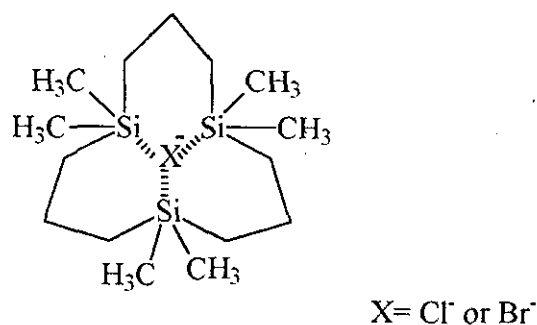


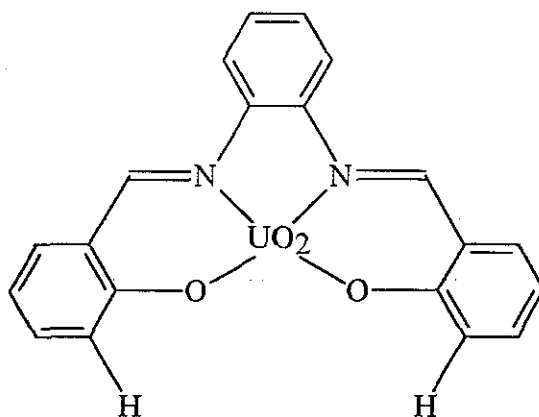
Fig. 9 Complexation of a halide anion by a crown silane

Research was carried out into the transport properties of these novel complexes. Jung and Xia set up several U-tube experiments using a saturated aqueous solution of tetramethylammonium halide in one arm, pure water in the other arm, and a methylene chloride solution of the crown silane in **Fig. 9** in the centre part of the U-tube which also had a magnetic stir-bar for mild mixing. It was reported that no transportation of fluoride or iodide ion was observed, while chloride and bromide were transported by the crown silane in **Fig. 9** from one aqueous solution to the other.

Uranyl cation

A Dutch research group led by Reinhoudt has been working on the incorporation of Lewis acidic uranyl cations into acyclic ligands.⁶⁴ They have previously reported that the uranyl cation makes excellent receptors for the complexation of neutral molecules due to co-ordination of a nucleophilic group ($\text{C}=\text{O}$, $\text{S}=\text{O}$, N) to the uranyl centre in addition to H-bond formation and aromatic interactions. The group studied the receptor molecules such as **19** on the addition of anions such as Cl^- and H_2PO_4^- . The crystal structure was solved, showing that the uranyl cation is coordinated to two oxygen atoms and two nitrogens atoms of the salophene unit and to the chloride anion,

which clearly showed anion complexation. It also clearly indicated complexation of the Cl^- anion by the uranyl cation.



19

A number of more selective anion receptors were prepared by the introduction of substituents which contain amido groups to the salene moiety.

1.3 Abiotic host molecules that contain a responsive function

Recent attention has been focused on a new generation of abiotic host molecules which contain responsive or signalling functions appended to or as an integral part of a host macrocyclic framework.⁶⁵ Illustrations of signalling functions include pH-responsive,⁶⁶ photochemical-responsive,⁶⁷ redox-responsive,⁶⁸ and temperature-responsive⁶⁹ where the receptors binding site strength and/or selectivity is influenced via the appropriate external physical or chemical trigger. The classes of interest in this research project are the redox-responsive and photochemical-responsive functions which will be reviewed in some detail.

Electrochemical recognition can take place due to a redox-centre being in close proximity to a host binding site.⁶⁸ Design of the signalling function for host molecules has two approaches, either recognition can take place by through-space electrostatic interaction or by electrostatic communication via various bond linkages between the receptor site and the redox centre (**Fig. 10**).

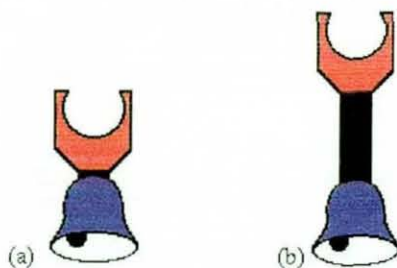


Fig. 10 Responsive receptors, the binding of a guest (G) is detectable:-

- a) through space interactions.
- b) through various bond linkages where the binding site is separated from the signalling unit by a bond or more.

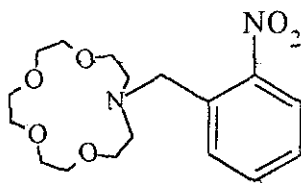
1.3.1 Electrochemical recognition of cations

Work has been completed into linking the electron-rich cavities of the macrocyclic polyethers, the crown ethers, and the three-dimensional cryptands with a variety of redox-active organic and inorganic centres with the intention of producing redox-responsive hosts which target alkali and alkaline earth metal cations.^{65, 68}

Sensing of alkali and alkaline earth metal cations by organic redox-active macrocycles

Gokel and co-workers were the pioneers in the field of electrochemical recognition. They made electrochemical observations on the addition of cations to crown ether containing or bearing reducible functions.⁷⁰ The effect of an addition of a cation to a crown ether with respect to a zero addition is that they may exhibit two redox waves or sometimes just a single redox wave that is shifted in position relative to the original redox couple.

Lariat ethers⁷⁰ were examined by Gokel and co-workers due to the presence of a side arm whose properties may be varied to control binding and selectivity. The Lariat ether **20** in the radical anion form enhances the binding strength of the cation compared to the unreduced form. This is due to the nitro function being reduced to the radical anion and thus forming a new type of intramolecular cation complex. It was discovered that binding was only enhanced when the nitro group was in the ortho position, rather than the para position. This is because the nitro function needs to be sterically positioned, so that it can pivot itself into position above the cation and the macrocyclic ring so that binding can occur. The addition of sub stoichiometric amounts of the guest metal cation to a solution of **20**, results in two well-resolved redox couples corresponding to the free ligand and the sodium cation complex. The second couple appeared on the addition of sub stoichiometric quantities of Na^+ , due to the electron-withdrawing ability of Na^+ which makes the nitroaromatic residue more easily reduced. The new redox couple is observed at a more positive potential.

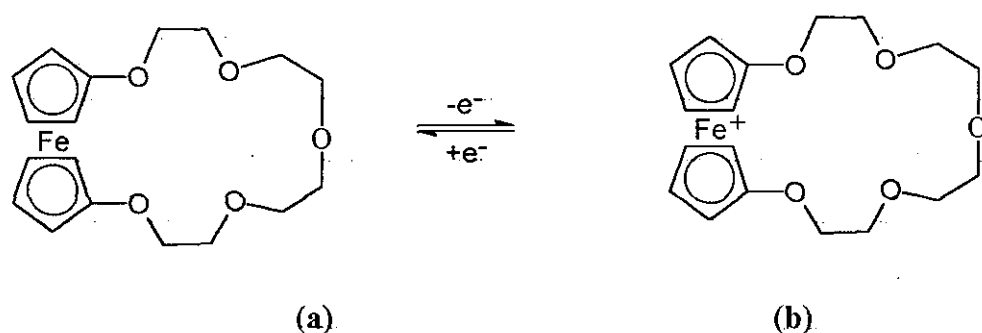


20

Redox-active macrocycles containing transition metal organometallic redox centres

Organometallic ferrocene moieties provide an attractive redox centre due to a well behaved reversible one-electron oxidation in most common solvents.⁷¹ The ferrocene can also be integrated into macrocyclic polyether ligands, which is made easier by its well established functional group chemistry.

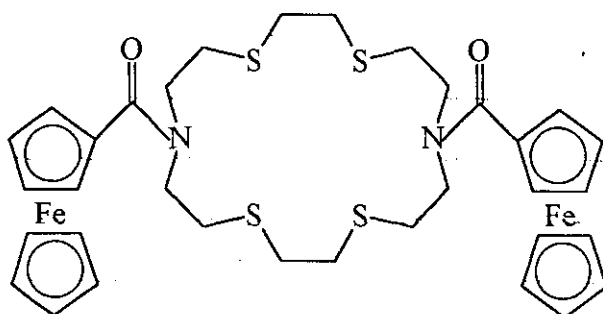
The first evidence of anodic shifts in the oxidation potential of ferrocene crown ethers was shown by Saji⁷² on pentaoxa[13]-ferrocenophane (**Scheme 9**) resulting from the addition of alkali metal salts. Two distinct electrochemical cyclic voltammetry (CV) waves were noted which corresponded to the complexed and uncomplexed (**b**) and (**a**). The complexed CV wave shows an anodic shift with respect to the uncomplexed (**a**) due to the binding constant of (**a**) decreasing compared to that of (**b**). In this case the decrease in the binding constant results from the mutual electrostatic repulsion between the ferricinium positive charge and the guest alkali metal cation. The magnitude of the shift in the anodic direction was found to be dependent upon the polarising power of the metal cation guest. The greater the polarising power of the metal the larger the decrease in binding constant, demonstrated by the fact that Li^+ produced a larger electrochemical shift than Na^+ .



Scheme 9 Redox processes of pentaoxa[13]-ferrocenophane

Electrochemical recognition of transition metal guest cations by ferrocene aza-, -thia-donor macrocyclic ligands

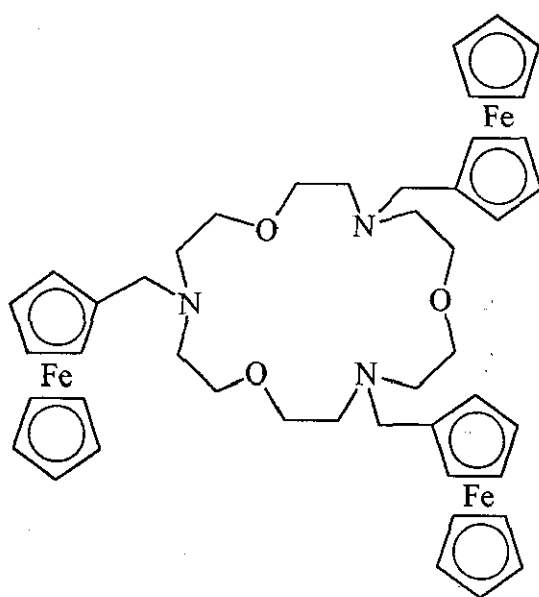
In addition to the ferrocene crown ethers ligands reported previously, a variety of related macrocyclic ferrocenophanes containing various heteroatoms such as sulphur, oxygen, and nitrogen have been synthesised⁷³⁻⁷⁷ which complex transition metal guest cations such as Ag(I), Cu(I), and Pd(II).^{78,79} Compound **21** shows an electrochemical shift of the ferrocene redox couple to a more positive potential on the co-ordination of the Cu(II) ion.



21

Ammonium cation electrochemical recognition

Production of a redox-active macrocycle⁸⁰ stems from 4,10,16-triaza-18-crown-6 **22** synthesised by Graf and Lehn.³⁹ This macrocycle forms three complementary $^+NH\cdots N$ hydrogen bonds between the host with NH_4^+ and primary ammonium guest complexes. On the incorporation of ferrocene redox-centres into the macrocycle, it was noted that on the addition of NH_4^+ to the electrochemical solution there was a significant one-wave anodic shift of the couple.



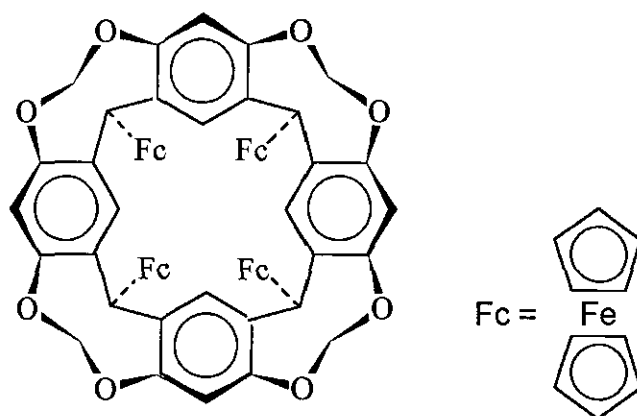
22

1.3.2 Electrochemical recognition of neutral guest species

Redox-active cavitand host molecules

Host molecules have been designed by using principles found in Nature's catalysts, the enzymes, which selectively bind and catalyse chemical reactions upon organic guest substrates. One such class of host molecules is the cavitands^{81, 82} which contain rigid hydrophobic cavities of dimensions large enough to include simple neutral organic guest

species. These receptors have been synthesised to incorporate redox-responsive functions in close proximity to the hydrophobic receptor cavity, with the intention of producing cavitands which can electrochemically recognise the inclusion of a neutral organic guest in the host cavity (Fig. 11). X-ray crystallographic studies were performed on a single crystal of **23**,^{83, 84} which showed that the cavitand was able to bind a single dichloromethane guest molecule within its host cavity. Molecular mechanics calculations on **22**⁸⁰ showed that it has the ability to complex small neutral molecules, thus acting as a neutral guest receptor. Cyclic voltammetry was performed on **23** which showed that the ferrocene redox-centre underwent reversible one-electron oxidation in an electrochemical solution of dimethylformamide, with no shift in the redox-couple being noted on the addition of dichloromethane. It was postulated with evidence from molecular mechanics calculations that the dichloromethane molecule did not penetrate far enough into the cavity for the redox-centres to be influenced. Also even if it did penetrate far enough in the cavity there would only be a small influence due to the neutral charge compared with the effect shown by alkali, alkaline earth metal and ammonium cations.



23

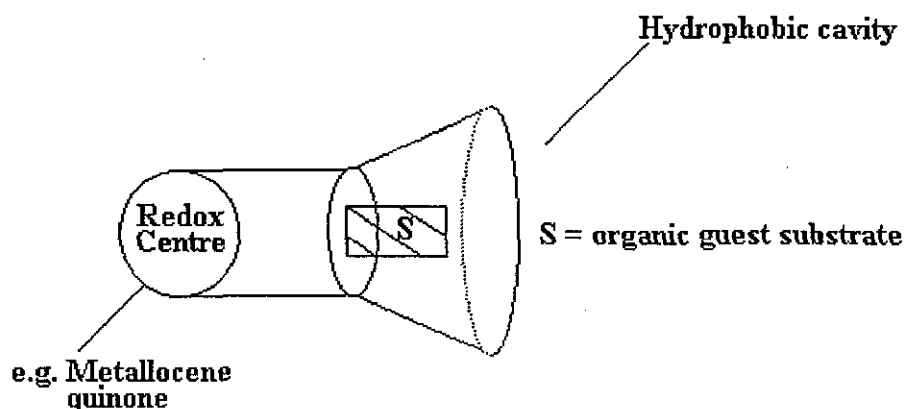


Fig. 11 Simple concept of electrochemical recognition: the binding of an organic guest (S) in close proximity to redox-active centres

1.3.3 Electrochemical recognition of anions

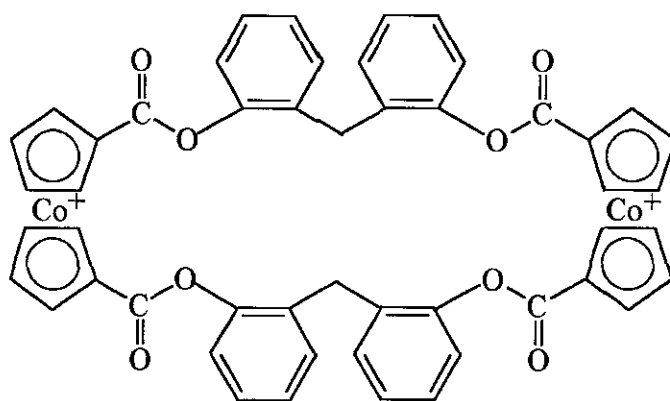
Molecular recognition of anion guest species by positively charged or electron-deficient neutral abiotic receptor molecules is a recent addition to the expanding field of electrochemical recognition.⁸⁵ This has been achieved by the development of the field of anion complexation and by the linking of redox centres to the host molecule.

There have been limited developments in the field of anion receptors with only a few classes of anion receptors being reported such as Lewis acid containing ligands,⁶⁰⁻⁶⁴ ammonium quaternary salts,³⁸ protonated polyamines⁴⁸ and guanidines.^{58, 59}

Macrocyclic and acyclic polycobaltocenium ligand systems

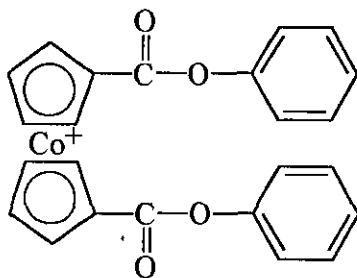
One of the first redox-responsive classes of anion receptor was reported by Beer and co-workers which incorporated the cobaltocenium moiety.^{86, 87} This resulted in the formation of a positively charged, pH-independent, air stable, 18-electron and redox-

active anion receptor. The cobalt acts as a Lewis-acid centre which favours the complexation of anions. They discovered that on addition of stoichiometric amounts of tetrabutylammonium bromide to an electrochemical solution of the hexafluorophosphate salt of **24** there was a gradual cathodic shift in the reversible reduction wave of the host.



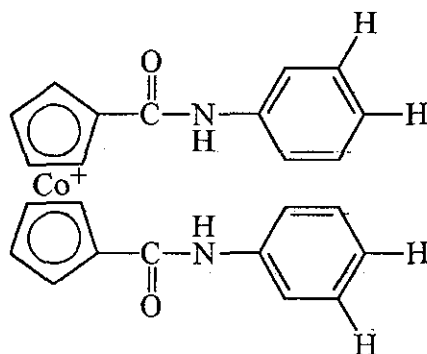
24

The same procedure was repeated with the hexafluorophosphate salt of **25** which exhibited no shift in the redox couple, thus implying that the complexation of the bromide anion within the macrocyclic cavity of **24** is critical for electrochemical detection.



25

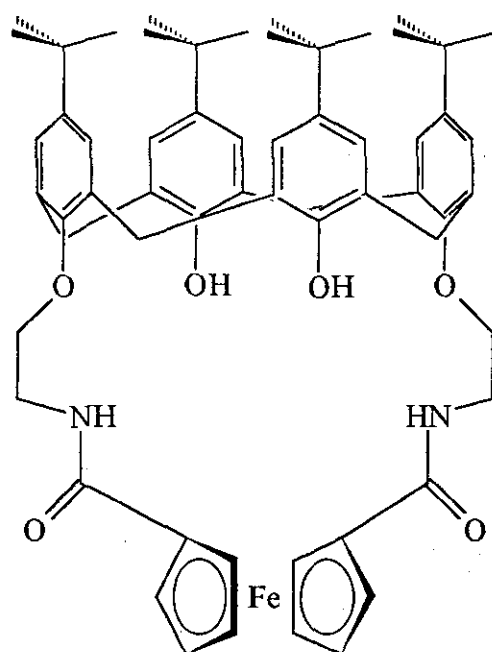
Due to the poor stability of the macrocyclic polycobalticinium, Beer and colleagues decided to use the stable amide linkages to construct novel acyclic cobaltocenium anion receptors such as **26**.⁸⁷ The results of ^1H NMR experiments suggest that there is significant $-\text{CO}-\text{NH}\cdots\text{anion}$ hydrogen bonding interaction, which contribute to the overall anion complexation process.



26

Neutral transition-metal Lewis-acidic recognition sites

The previous cobaltocenium receptors underline the importance of hydrogen bonding in the anion recognition process.⁸⁸ It was therefore reasoned that the neutral Lewis acidic centre ferrocene could be incorporated instead of the cobaltocenium.⁸⁹ This led to the synthesis of ferrocene containing complexes such as **27**. This complex binds anions due to the combination of the favourable Lewis acid-anion electrostatic and amide ($\text{CO}-\text{NH}$) anion hydrogen-bonding interaction. The neutral complex **27** shows a significant cathodic perturbation on the addition of anionic species. In fact the complex is capable of detecting the H_2PO_4^- anion in the presence of ten fold excess amounts of HSO_4^- and Cl^- ions.⁸⁸



27

A number of water soluble polyaza ferrocene macrocycles such as the complexes in **Fig. 12**⁸⁹ have been shown to bind phosphate anions (H_2PO_4^- , ATP) in the pH range 6-7.

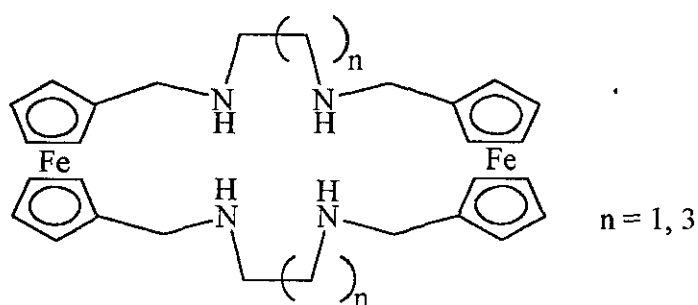


Fig. 12 Water soluble polyaza ferrocene macrocycle.

1.3.4 Photoresponsive - Fluorophores

Fluorescence spectroscopy is increasingly becoming an important tool in the study of Host-Guest complexes.⁹⁰ The attraction of fluorimetry lies in its high sensitivity and high selectivity. A number of molecules have been designed to incorporate fluorophores. On the incorporation of a fluorophores it has been noted that a number of host molecules undergo a perturbation in their fluorescence emission spectrum on complexation with a guest. Perturbations in the fluorescence emission spectrum may manifest as a change in either intensity or a shift in λ_{max} or both. Complexation can cause a perturbation in the fluorescence emission spectrum by affecting:-

1. Quantum yields:- fluorescence, phosphorescence and the lifetimes of the excited states may be increased or decreased, resulting in an increase or decrease in the intensity of the spectrum.
2. Energy levels:- change in the energy levels on complexation which causes a perturbation in the spectrum to higher or lower energy.

1.3.5 Photochemical recognition of cations

Crown ether fluorophores

A number of crown ether fluorophores have been investigated which show binding of alkali metal cations and perturbation in the fluorescence-emission spectra. Sousa and Larson^{91, 92} produced a number of crown ether naphthalene derivatives which utilised the naphthalene chromophore and monitored its response to a variety of perturbors (Fig. 13 and 14). They discovered that the perturbation of the fluorescence emission was dependent on the orientation of the chromophore and on the complexed perturber. The compound in Fig. 13 on complexation with alkali metal cations showed a decrease in fluorescence quantum yield, an increase in phosphorescence quantum yield, a slight decrease in phosphorescence lifetime as well as a small blue shift in fluorescence. The crown ether naphthalene derivative in Fig. 14 however showed an increase in

fluorescence quantum yield, a decrease in phosphorescence quantum yield, a substantial decrease in phosphorescence lifetime and a red shift in fluorescence.

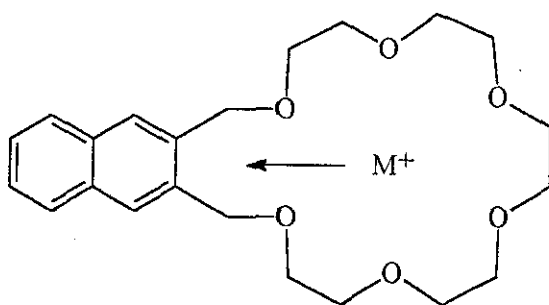


Fig. 13 Crown ether naphthalene derivative

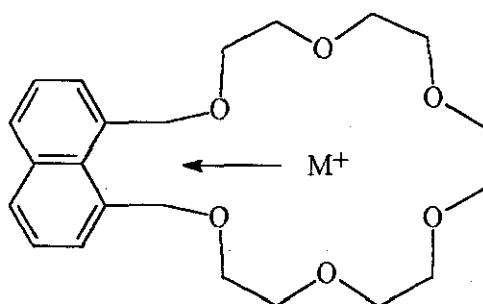


Fig. 14 Crown ether naphthalene derivative

Perturbation of the photoexcited states by alkali metal cations in this case appear to be attributed to a complexation-induced change in triplet energy relative to ground and excited singlet state energies. Sousa and Larson proposed that the direction dependence of the alkali metal cation effect may be due to:-

1. Direct cation-chromophore interaction.
2. Conformational changes in the chromophore enforced by crown-metal complexation.
3. Conformational changes in the chromophore enforced by solvent orientation about the complexed cation.

Macrobicyclic anthraceno-cryptands

Lehn and co-workers⁹³ synthesised the receptors in **Fig. 15** so that they could combine the specific complexing ability of the cryptands with the photophysical behaviour of the anthracene ring.

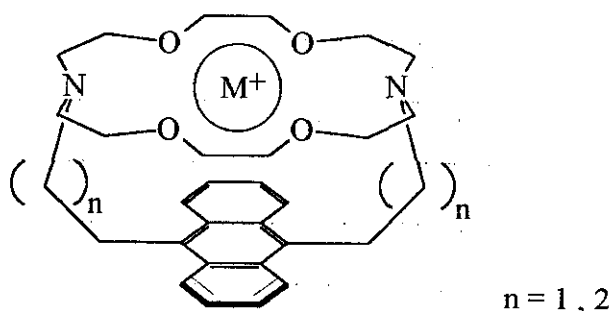


Fig. 15

This system was designed because they expected that the combination of a strong ligand such as a cryptand and an aromatic chromophore would lead to pronounced photophysical responses in the presence of the salt. The fluorescence of the receptor in **Fig. 15** showed significant changes in intensity and λ_{max} on the addition of cations such as K^+ and Ag^+ .

1.3.6 Photochemical recognition of neutral guest species

Shinkai et al.⁹⁴ have synthesised a very interesting receptor for the molecular-recognition of neutral molecules. This molecule (**Fig. 16**) undergoes a number of photochemical processes:-

1. Intramolecular quenching - the pyridine unit quenches the fluorescence from the pyrene unit.
2. Monomer emission - pyrene unit fluoresces at 378 nm when excited at 345 nm.
3. Excimer emission - the two pyrene units undergo excimer emission at 480 nm.

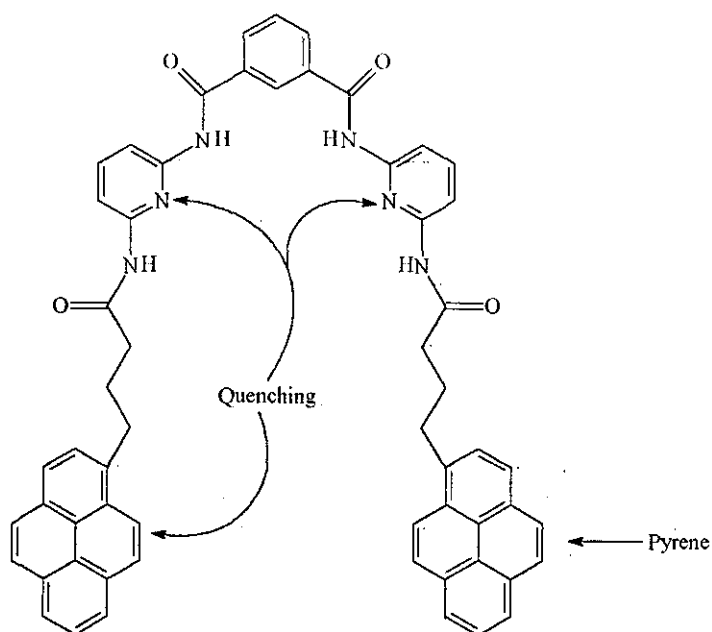


Fig. 16

Complexation of the neutral molecule occurs due to the formation of hydrogen bonds between the pyridine and a number of organic guests such as barbitol (**Fig. 17**). The incorporation of such a guest leads to a change in the photochemical properties of the host:-

1. Intramolecular quenching between the pyrene and pyridine decreases.
2. Decrease in intramolecular quenching leads to an increase in monomer emission.
3. Excimer emission decreases due to steric effects.

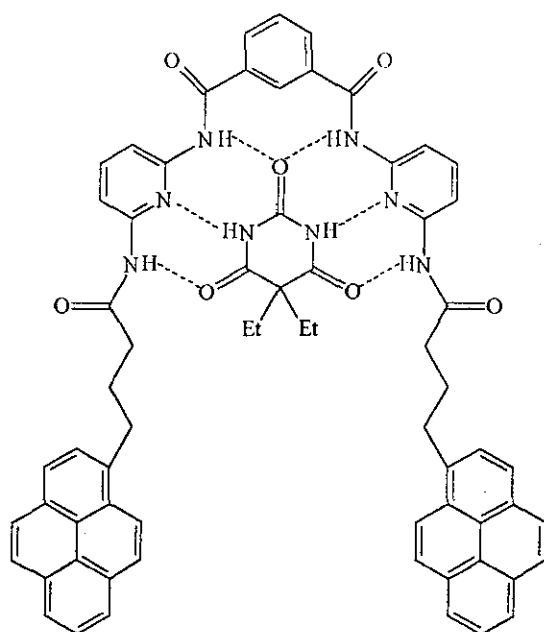
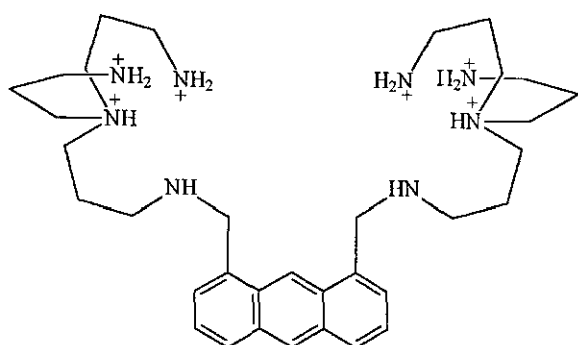


Fig. 17

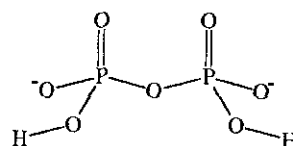
1.3.7 Photochemical recognition of anions

The design and syntheses of specific receptors which have the capability of optically detecting anions in aqueous or non-aqueous media are extremely rare. Czarnik and Vance⁹⁵ have recently described the only anion fluorescence responsive type, based on acyclic anthracene appended polyammonium receptors. The fluorescence receptor **28** binds pyrophosphate **29** over 2200 times more tightly than phosphate at pH 7, permitting the real-time monitoring of pyrophosphate hydrolysis. In the uncomplexed form receptor **28** has a low fluorescence intensity and the guest **29** is not fluorescent. However on complexation the receptor molecule becomes highly fluorescent. This is thought to be due to enhanced fluorescence as a result of intracomplex amine protonation.



28

low fluorescence intensity

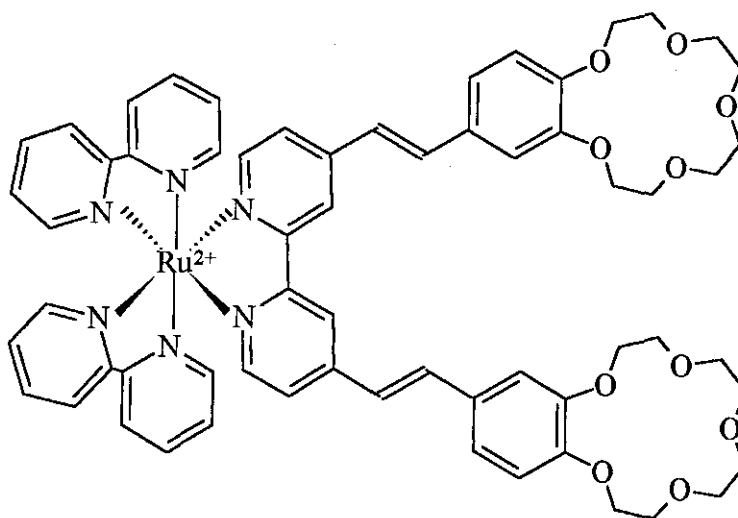


29

non-fluorescent

Ruthenium(II) trisbipyridyl derivatives

A novel group of functionalised crown ether such as receptor **30** demonstrating spectrochemical recognition of alkali and alkaline earth cations were reported by Beer and colleagues.^{96, 97} The receptor sites were linked to a trisbipyridyl ruthenium (II) moiety which acted as the fluorophore. Trisbipyridyl ruthenium (II) and its derivatives are well documented and have a predictable photochemistry.



30

Ruthenium(II) polypyridyl complexes such as **30** exhibit metal-to-ligand charge transfer (MLCT) band for the electronic absorption and fluorescence spectra.⁹⁷ Fluorescence measurements of the MLCT bands of these complexes provides a useful tool to assess the effect of different ligand, as well as probing metal-cation binding on excited-state energies.

The MLCT emission maxima for the reported ruthenium(II) complexes showed a significant shift to lower wavelengths and an increase in intensity in the presence of Na⁺, K⁺ and Mg²⁺. Binding of metal cations in the benzocrown ether will prevent electron donation to the phenyl group, thus producing a shift of the m.l.c.t. band back to higher energies.

1.4 Aims of the PhD

The aim of this project is to extend the field of molecular recognition of anions and cations of biochemical, medical, chemical and environmental importance. This will be achieved by the use of a number of novel receptor molecules that are designed to bind anionic and cationic guests. The binding of the guest anions and cations will be probed by various electrochemical, spectrochemical and ¹H NMR spectroscopy techniques. The receptor molecules to be studied will include (i) ruthenium(II) trisbipyridyl complexes of acyclic, calix[4]arene and cyclic 2,2'-bipyridine ligands, (ii) the macrocycle N-phenylaza-15-crown-5, and (iii) crown ether derivatives of diquat. The overall aim is to develop a spectrochemical and/or electrochemical sensor for anions and cations.

Chapter 2 - Experimental

2.0 Instrumentation and procedures

2.0.1 Electrochemistry

Electrochemical measurements were conducted using an EG&G Princeton Applied Research (PAR) 173 potentiostat with a PAR 175 universal programmer and a PAR 179 digital coulometer. A three-compartment electrochemical cell was employed which was built 'in house' (**Fig. 1**), with a platinum wire (0.32-cm^2 surface area) or indium-tin oxide (Balzers, $20\Omega/\square$) optically transparent conducting working glass electrodes for cyclic voltammetry.

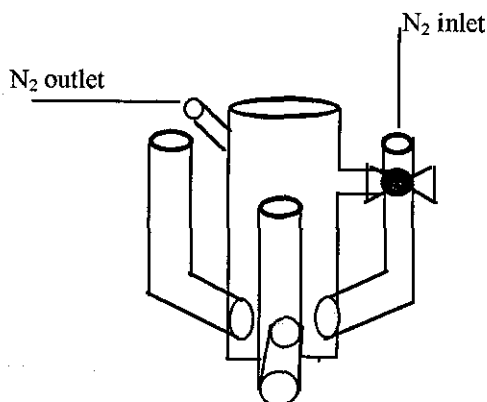


Fig. 1 Three compartment electrochemical cell

A platinum mesh working electrode was used for constant potential electrolyses, in all cases a platinum-mesh counter electrode was utilised. Electrode potentials were measured and are quoted with respect to a Radiometer sodium chloride saturated calomel electrode (SSCE) at $25\pm 2^\circ\text{C}$. No iR compensation was employed. Both counter and reference electrodes were each separated from the working electrode

compartment of the electrochemical cell by glass frits. Solutions undergoing constant potential electrolyses were magnetically stirred throughout to enhance mass transport. Platinum working electrodes were pre-treated by immersion in concentrated sulphuric acid; anodisation, then cathodisation, (2 min each at 100 mA in 0.5 mol dm^{-3} sulphuric acid) followed by washing with de-ionised water, then acetonitrile and finally air dried. Electropolymerisation investigations involved potential cycling through the ligand-centred reductions to activate the vinylic linkages. The advantage of this procedure compared with electroreduction at constant negative potentials is that the slower growth rate leads to more uniform films.⁹⁸ For successful electropolymerisations, the resulting polymer modified electrodes were rinsed in acetonitrile prior to examination in pure supporting electrolyte solutions. All electropolymerisation profiles and subsequent polymer redox responses reported were reproducible. Measurements were carried out in deoxygenated acetonitriles by purging the solution with solvent saturated N_2 .

Theory

Cyclic voltammetry is a popular and powerful technique for the initial studies of new systems.⁹⁹⁻¹⁰¹ The technique is also very rapid allowing many experiments to be carried out within a few minutes. In this technique, the applied potential is varied with time in a symmetrical saw-tooth waveform, while the resulting current is recorded over the entire cycle of the forward and reverse sweeps. Cyclic voltammetry can provide information on the :-

1. reversibility of an oxidation/reduction process
2. determination of mechanisms and identification of intermediates

The above information can be obtained by varying a number of experimental conditions:-

1. potential scan rate which is defined as the rate of change of potential with respect to time
2. potential limits E_{initial} , E_{limit} and E_{final} and direction of the sweep

The reversibility of system can be determined by a trained eye but it is also important to compare the experimental result with the theoretical response (**Table 1**) and the reversible potassium ferricyanide system (**CV 1**).

- $\Delta E_p = E_p^A - E_p^C = 59/n \text{ mV}$
- $E_p - E_{p/2} = 59/n \text{ mV}$
- $-j_p^C/j_p^A = 1$ (note j_p^C and j_p^A are of opposite sign)
- $j_p \propto \nu^{1/2}$
- E_p are independent of ν
- at potentials beyond E_p , $j^{-2} \propto t$

E_p = peak potential (A=Anodic and C=Cathodic) j_p = peak current

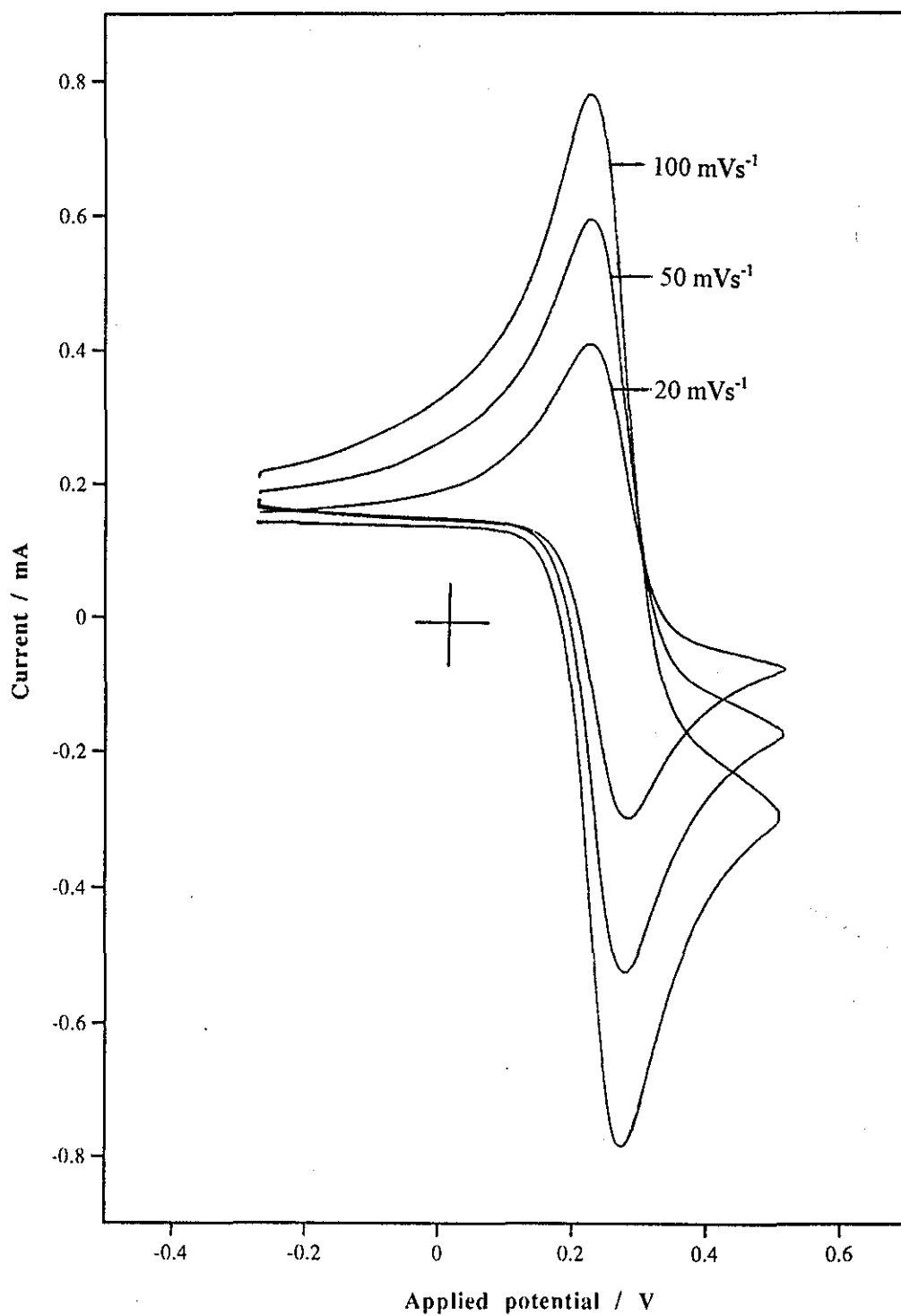
ν = scan rate

t = time

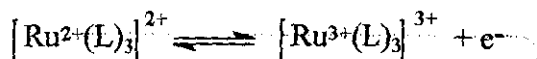
Table 1 Diagnostic tests for the form of the cyclic voltammetric response for a reversible electron transfer process at 298 K.

Electrochemical studies of ruthenium (II) trisbipyridyl are generally performed in non-aqueous aprotic solvents using cyclic voltammetry. The cyclic voltammograms of

CV1 - CVs at 20, 50 and 100 mVs^{-1} in an aqueous solution containing 10 mM potassium ferricyanide in 1 M potassium nitrate. Potentials given versus saturated calomel reference electrode.



[Ru(bipy)₃][PF₆]₂ and ruthenium(II) derivatives typically feature the reversible oxidation of the Ru²⁺ metal centred orbital to form the genuine Ru³⁺ complexes as shown in **Scheme 1**.¹⁰²



Scheme 1 Reversible oxidation of Ru²⁺ to Ru³⁺

Reduction of ruthenium(II) trisbipyridyl complexes is not as straight forward as oxidation and may involve either a metal-centred or ligand-centred orbital. Depending on the ligand field or the reducibility of the ligand, if the ligands are easily reduced then the reduction is ligand based. Three reversible waves are generally observed in acetonitrile (**Scheme 2**).



L = Ligand

Scheme 2 Three reversible reductions

2.0.2 Spectroscopy

A Hewlett-Packard HP 8452A diode-array spectrophotometer was employed for recording electronic absorption spectra. A PC-controlled Spex Model Fluoromax fluorescence spectrometer was used for recording the fluorescence (uncorrected)

emission spectra, the appropriate excitation wavelength corresponding to the λ_{max} of the metal-to-ligand charge-transfer (MLCT) absorption band being determined from the electronic absorption spectra. All measurements were conducted at $25 \pm 2^\circ\text{C}$ using a 1 x 1 cm rectangular quartz cuvette and deoxygenated acetonitrile solutions. Quantum yields (Φ) were calculated using the following equation.¹⁰³

$$\frac{\text{Fluorescence area 2}}{\text{Fluorescence area 1}} = \frac{\Phi_2}{\Phi_1} \times \frac{\text{optical density of 2}}{\text{optical density of 1}}$$

For each sample measurement, $[\text{Ru}(\text{bipy})_3][\text{PF}_6]_2$ in aerated water was used as standard ($\Phi = 0.028$).¹⁰⁴ The assumption is made that Φ will be independent of the anion of the salt; the chloride salt is used in this reference. This seems reasonable, as addition of chloride does not change Φ in DMSO. Nuclear magnetic resonance (NMR) spectra were obtained on a Bruker 400 instrument using the solvent deuterium oxide signal as internal reference.

Theory of the electronic absorption and emission of ruthenium(II) trisbipyridyl

Ruthenium(II) trisbipyridyl complexes exhibit the following excited states:- metal-centred (MC) excited states, which is derived from electronic transitions between σ to σ^* orbitals that are localised on the metal; ligand-centred (LC) excited states, which are derived from electronic transitions between π_T to π_T^* orbitals that are localised on the ligands; MLCT excited states which derive from electronic transitions between π_M to

π_T^* orbitals of different localisation.¹⁰² A broad low-energy MLCT emission band is observed due to absorption in the visible region by the MLCT band.

2.0.3 Chemicals and reagents

Acetonitrile (Fisons, AR), Acetonitrile- d_3 (Fluka) and spectroscopy grade DMSO (BDH) were used as received. All measurements were carried out in de-oxygenated acetonitrile (Fisons AR, stored over molecular sieves) and 0.1 M tetrabutylammonium tetrafluoroborate (Fluka, vacuum-dried and stored in a desiccator over silica gel) as supporting electrolyte. Cation complexation studies used the following chemicals without further purification N-phenylaza-15-crown-5:- (Merck), N,N-dimethylaniline (Lancaster), magnesium perchlorate (Aldrich), lithium perchlorate (Aldrich) and sodium perchlorate (Aldrich). (4-formyl-benzo)aza-15-crown-5 was prepared by Dr. O. Kocian (University of Birmingham).

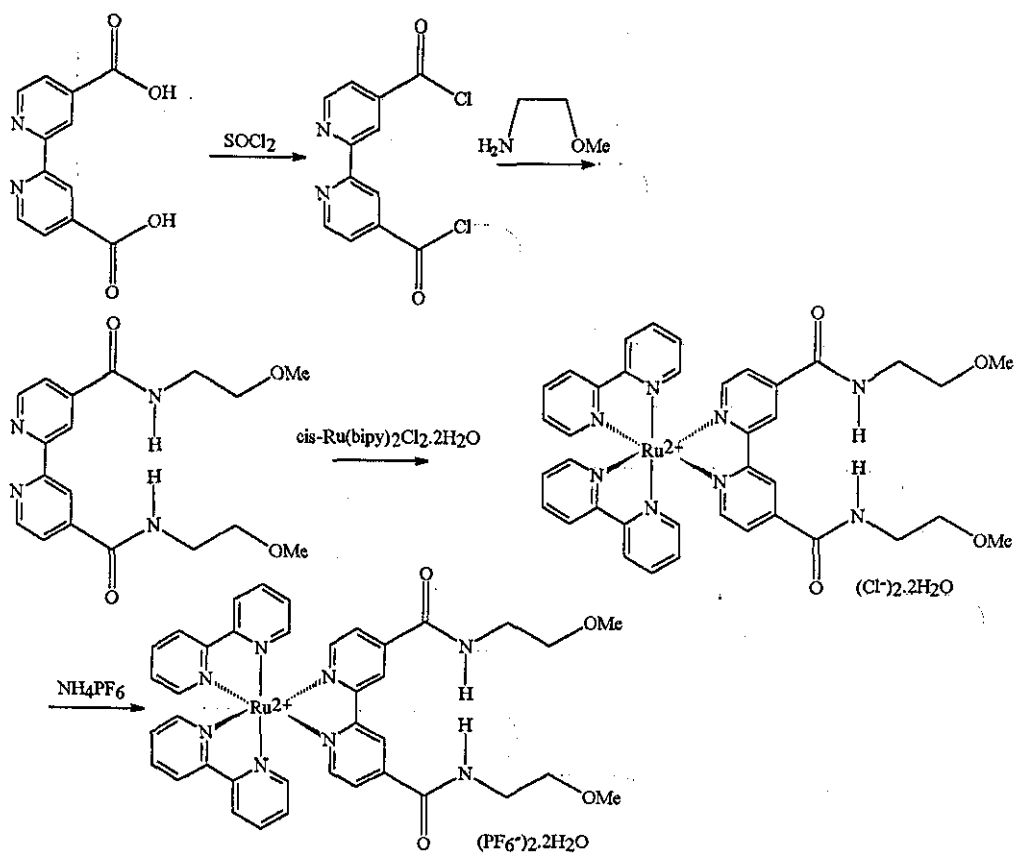
Tetrabutylammonium chloride hydrate (Aldrich), tetrabutylammonium bromide hydrate (Aldrich) and tetrabutylammonium dihydrogen phosphate (Aldrich) were employed in anion complexation studies and were used as received.

2.1 Synthesis

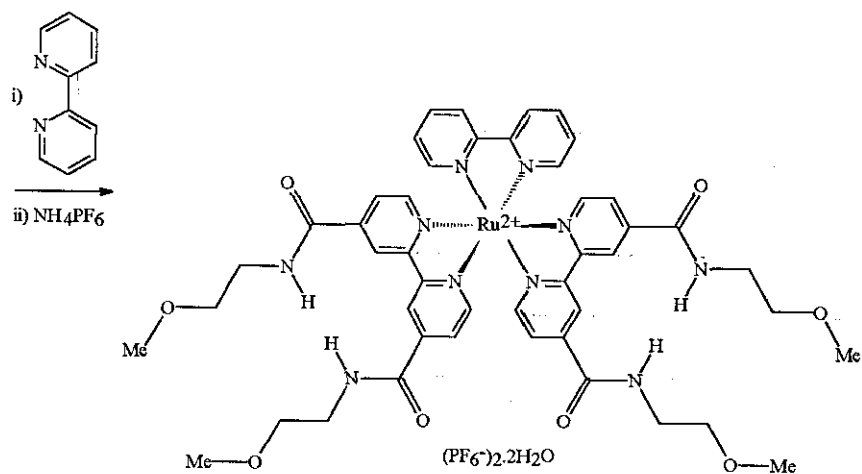
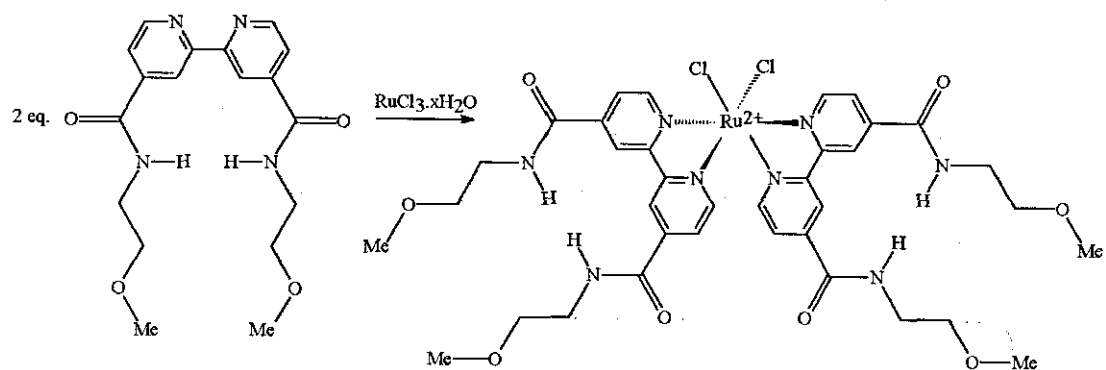
The following schemes detail the synthesis of the receptors studied in this thesis, which were prepared by Dr. Fridrich Szemes (Oxford University), Dr. Dusan Hesek (Oxford

University) and Dr. Oldrich Kocian (University of Birmingham). These receptor molecules were characterised using standard techniques including mass spectrometry and NMR spectroscopy.

Acyclic Ruthenium Bipyridyl Receptors investigated in Chapter 3

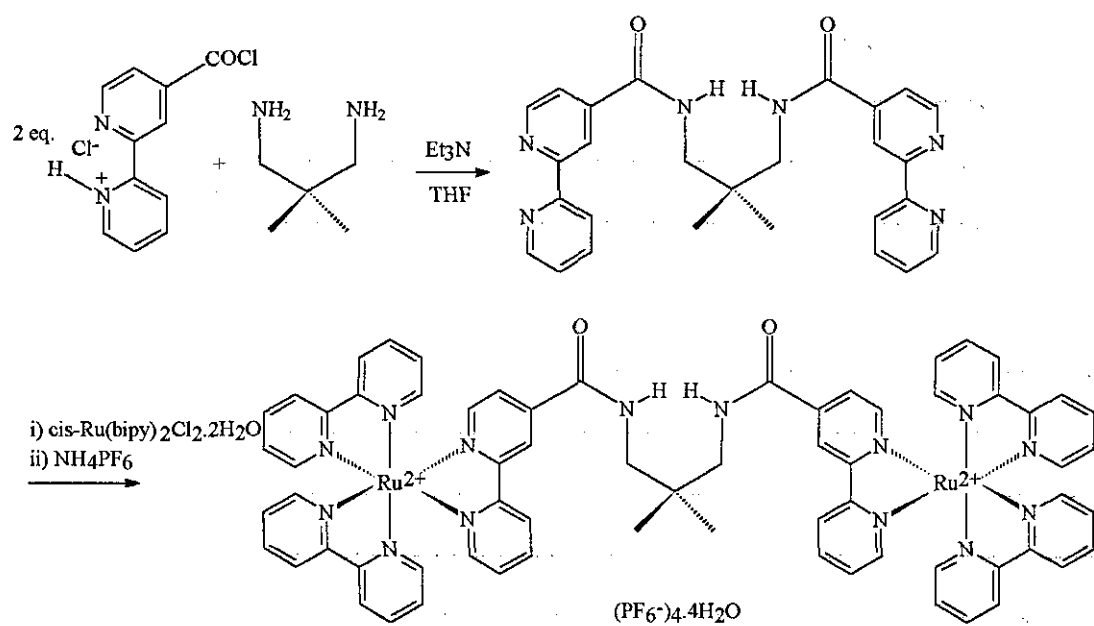


Complexes 1 and 2¹⁰⁵

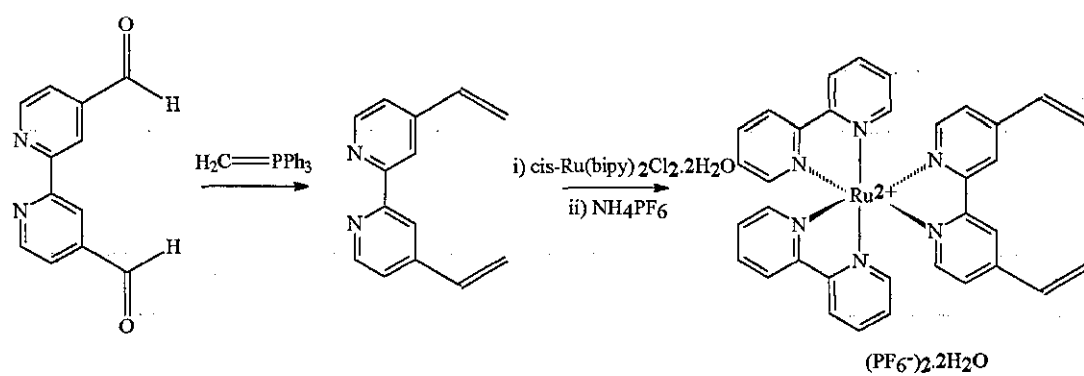


Complex 3

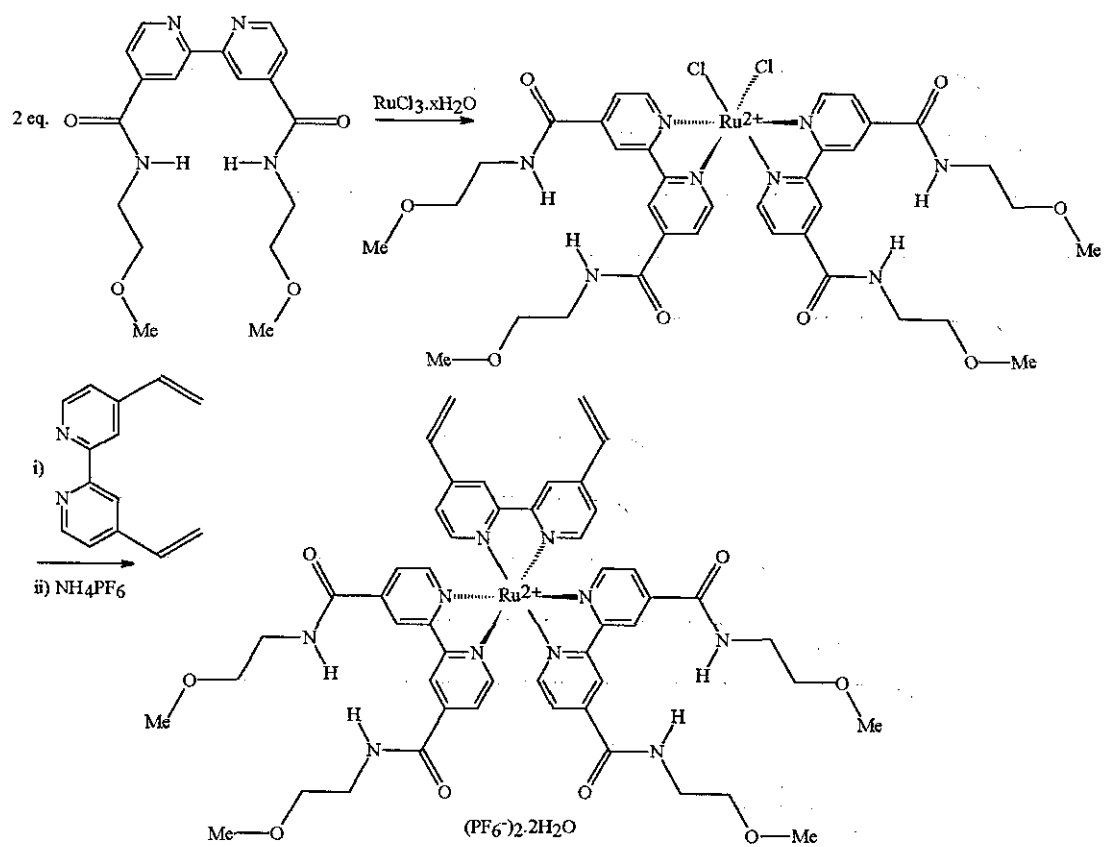




Complex 5

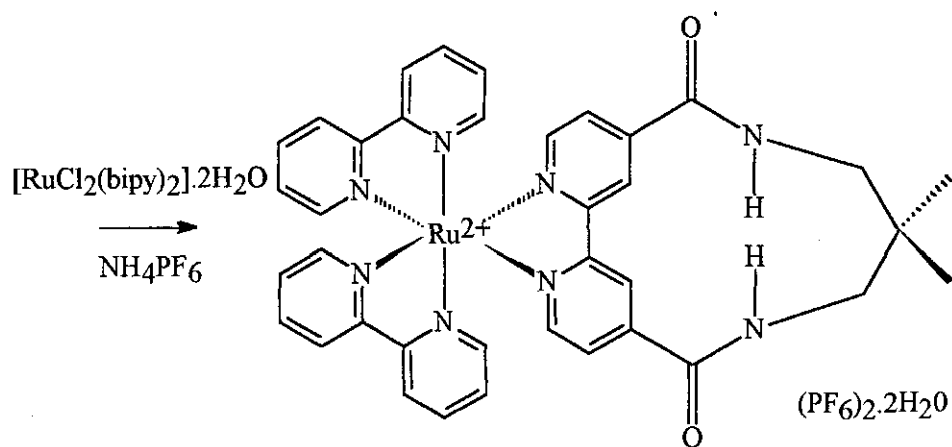
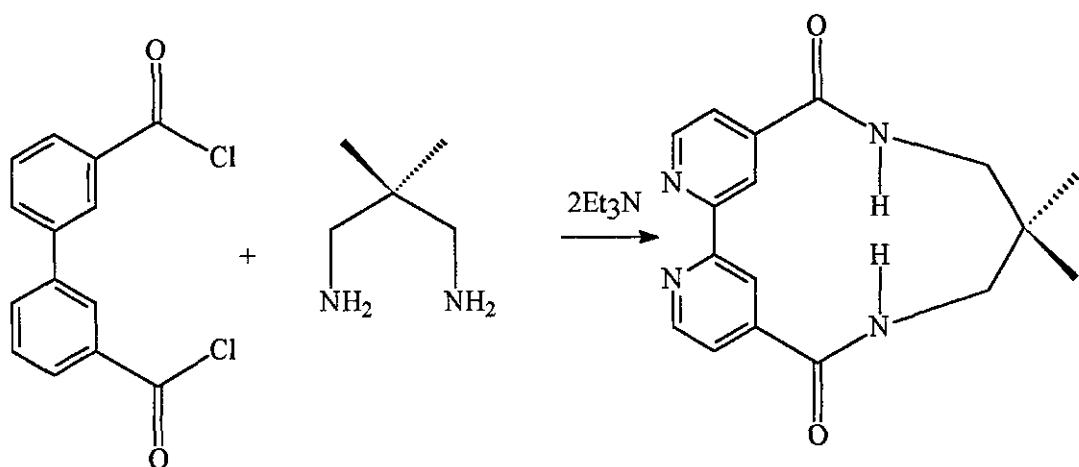


Complex 6

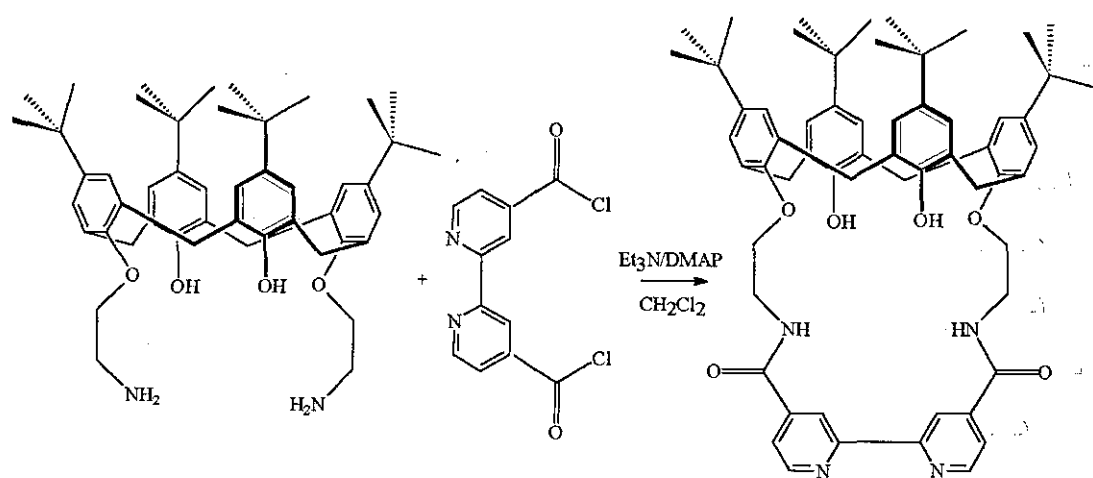


Complex 7

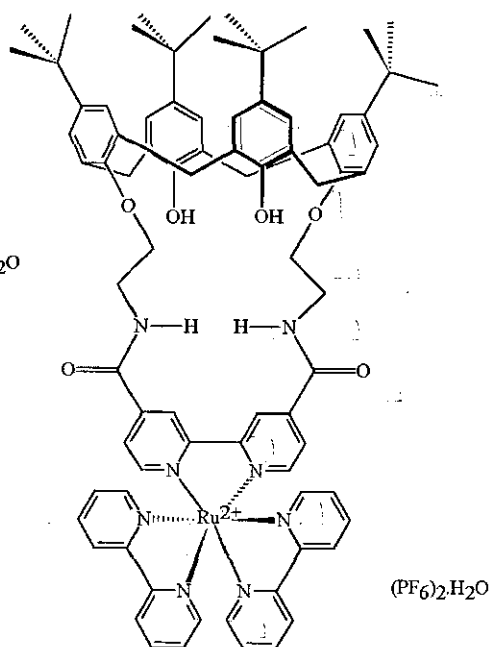
Macrocyclic and Calix[4]arene Ruthenium (II) Bipyridyl Receptors investigated in Chapter 4



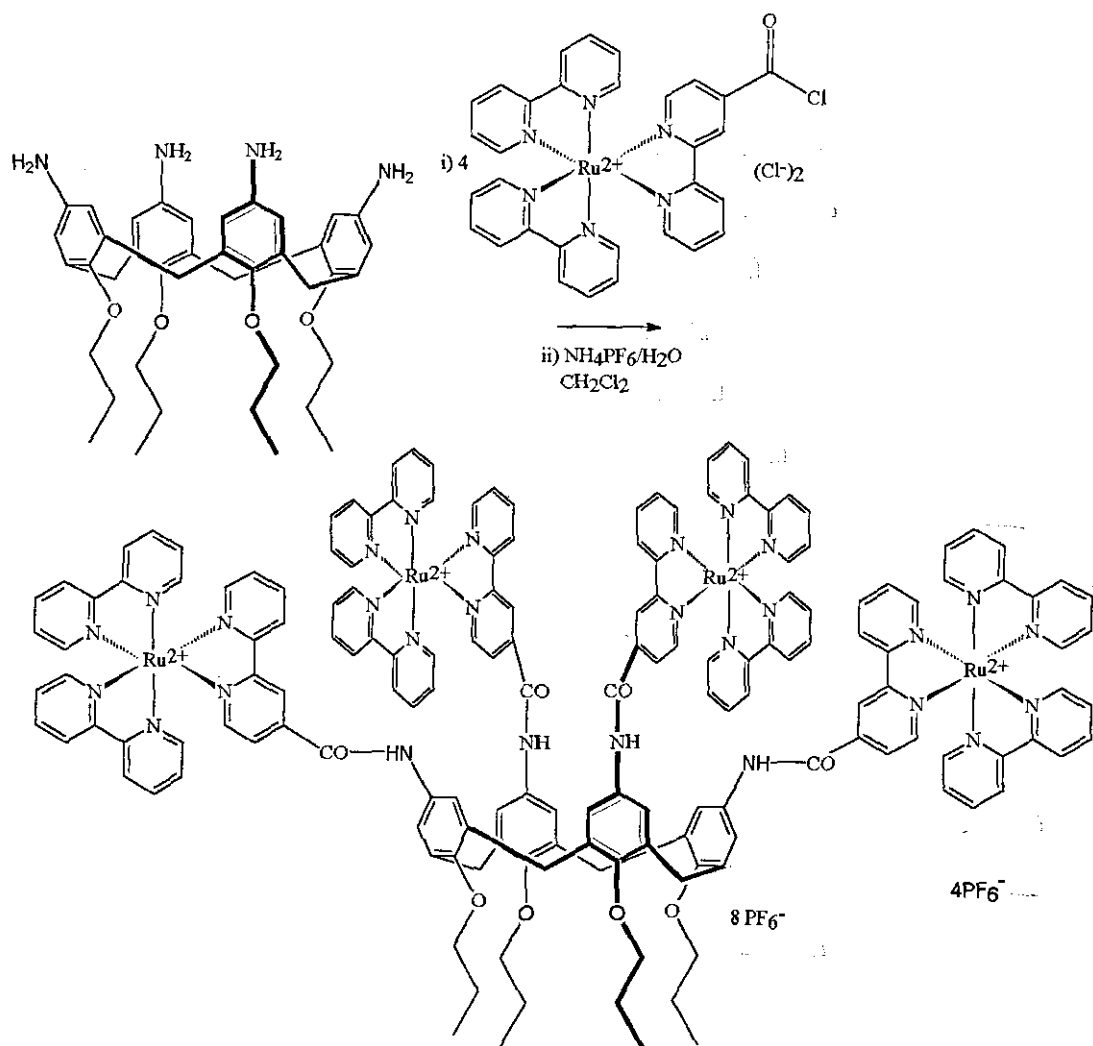
Complex 1¹⁰⁷



(i) $(\text{bipy})_2\text{RuCl}_2 \cdot 2\text{H}_2\text{O}$
(ii) NH_4PF_6

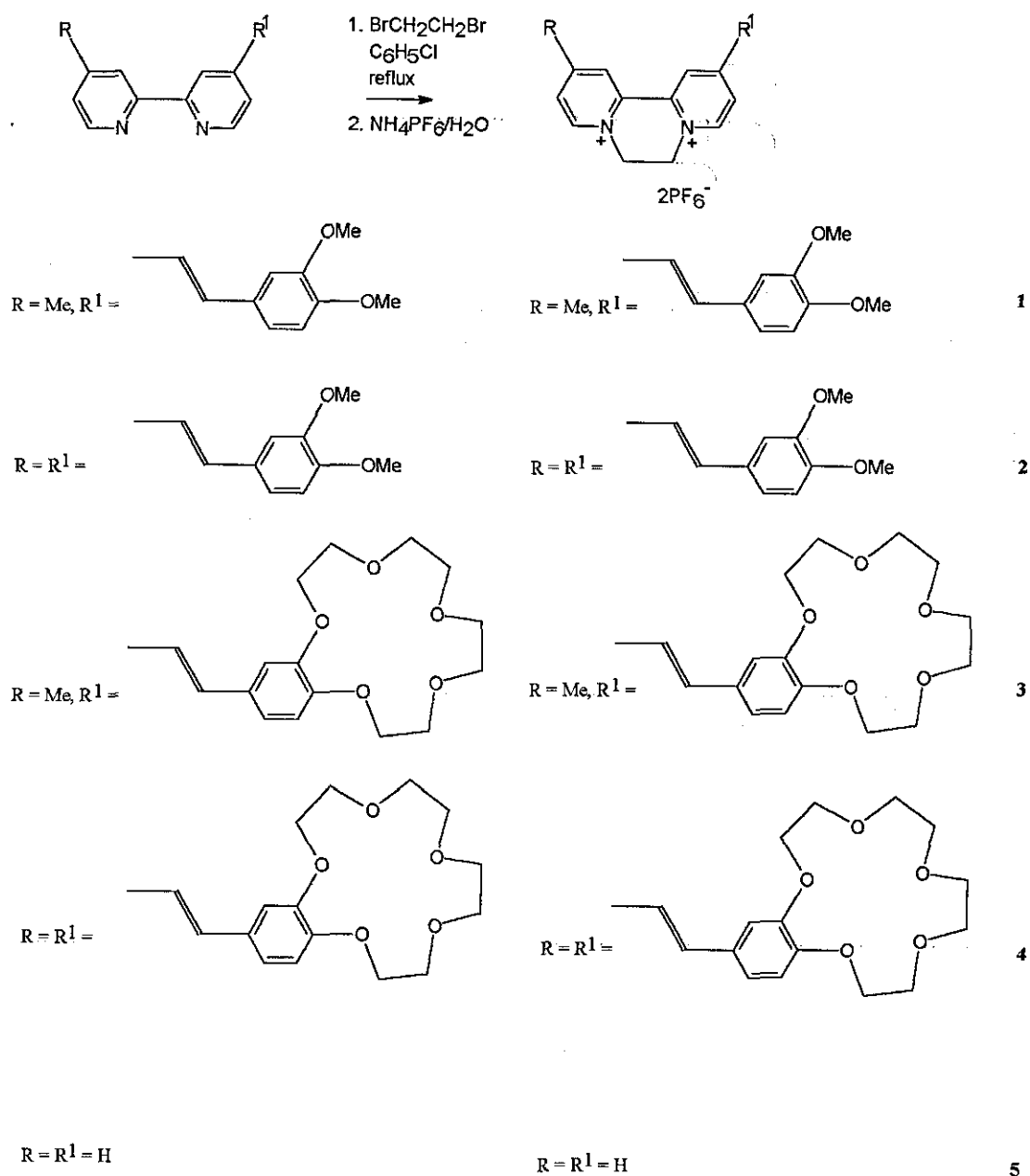


Complex 2¹⁰⁷

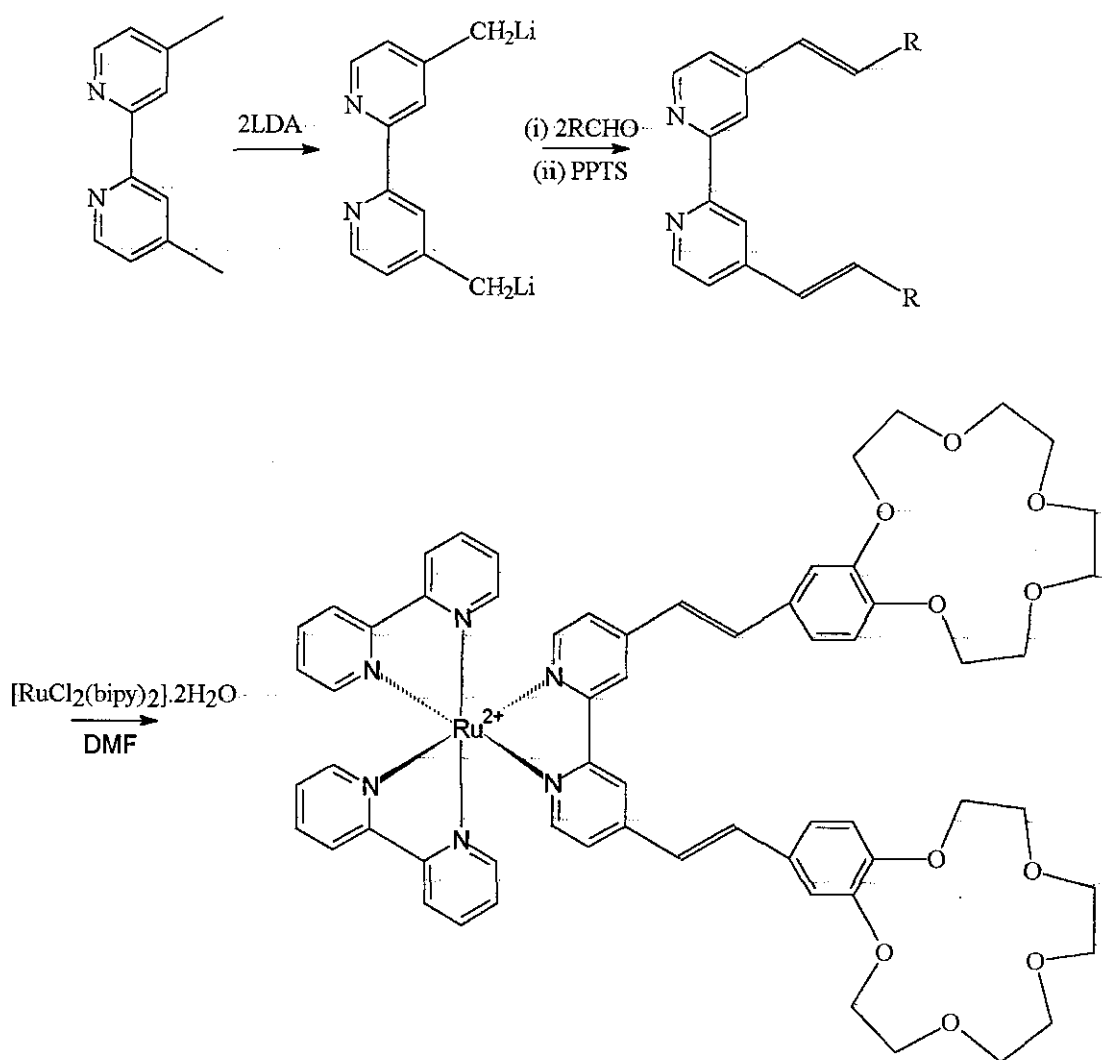


Complex 3⁸⁵

Diquat Receptors investigated in Chapter 5



Receptors 1 - 5



Receptor 6^{108, 109}

Chapter 3 - Acyclic Ruthenium(II) Bipyridyl Receptor Molecules

3.0 Introduction

Previous work by Beer and co-workers has underlined the importance of the hydrogen bond in the complexation of anions, where 4,4'-disubstituted-2,2'-bipyridines incorporating amide linkages proved to be good receptors.⁸⁵ It was also discovered that the coordination of the receptors to Lewis acidic transition metals (in particular Ru(II)) significantly enhanced their complexation ability. The attachment of the bipyridyl ligands to a metal centre such as ruthenium(II) provides a positive charge to attract the anion. This is due to the ruthenium(II) metal centre having a formal charge of +2 which can act in conjunction with the amide on the bipyridyl to bind anions. The ability of the receptor site to bind anions can be altered by varying the electron donating/withdrawing groups attached to the amide groups. This affects the acidity of the amide protons and its ability to form hydrogen bonds with anions (Fig. 1). Also, the ruthenium(II) trisbipyridyl is a redox and photo-active centre,¹⁰² hence providing a spectroscopic and electrochemical means of investigating the binding process.

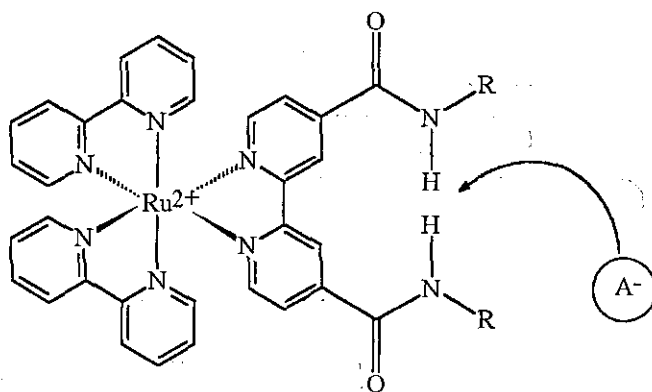
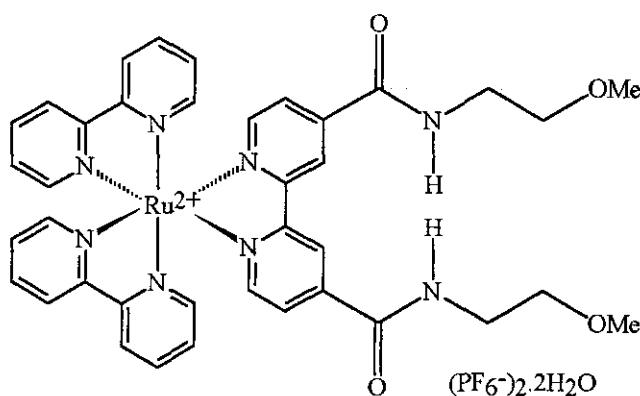


Fig. 1 Schematic representation of the proposed anion binding site

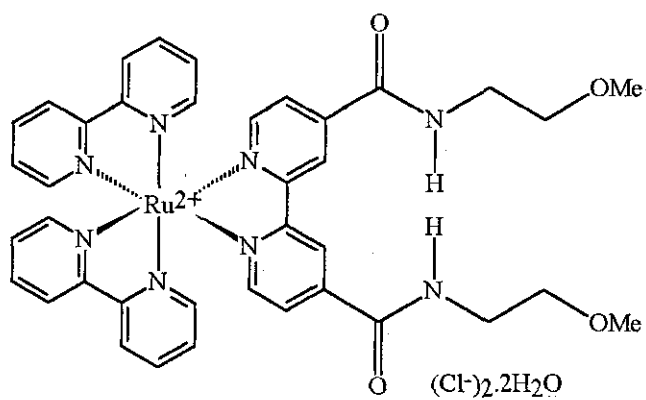
When R is electron withdrawing, the acidity of the amide protons increases and thus its ability to bind anions. However, an electron donating substituent has the opposite effect. The receptor backbone precursor 4,4'-dimethyl-2,2'-bipyridine was chosen for a number of reasons:-

1. it is commercially available
2. substitution at the 4-positions produces an acyclic ligand with a pseudo cavity which should be sufficiently large to envelope most anions

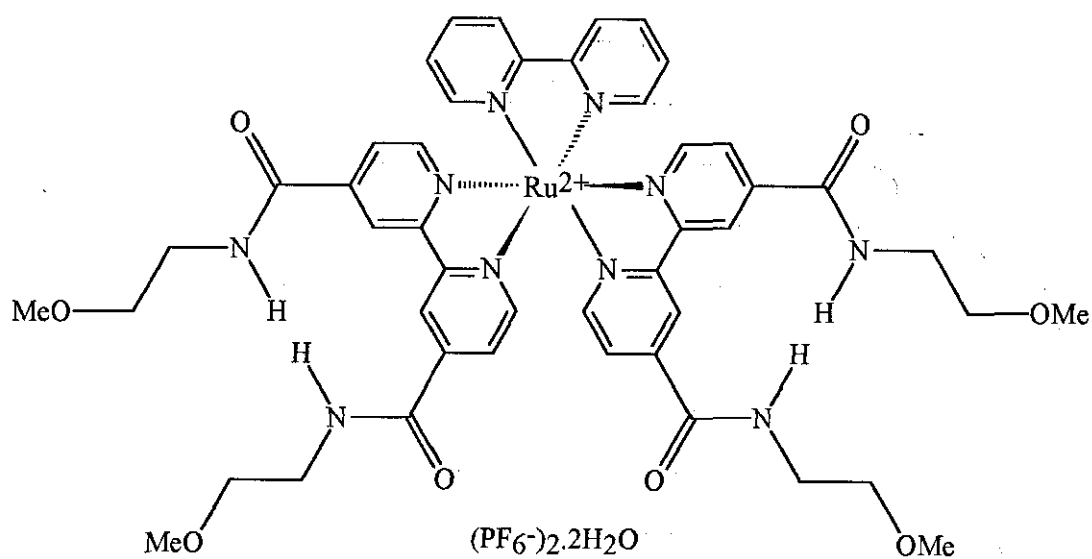
The ruthenium(II) trisbipyridyl acyclic receptor molecules **1-5** were synthesised (Chapter 2 - experimental) and characterised by Fridrich Szemes.¹⁰⁵⁻¹⁰⁷ Such that the receptors would provide a acyclic binding site for anions.



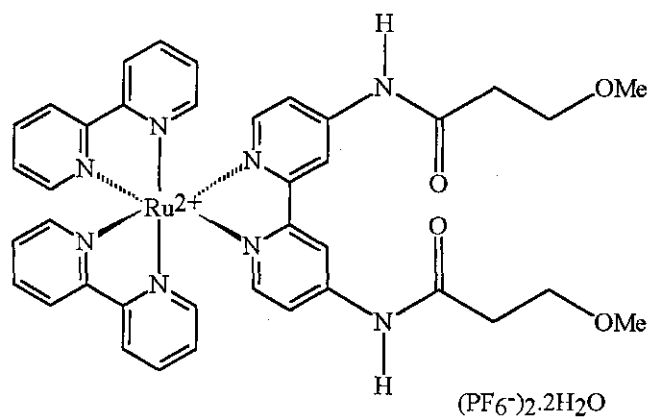
1



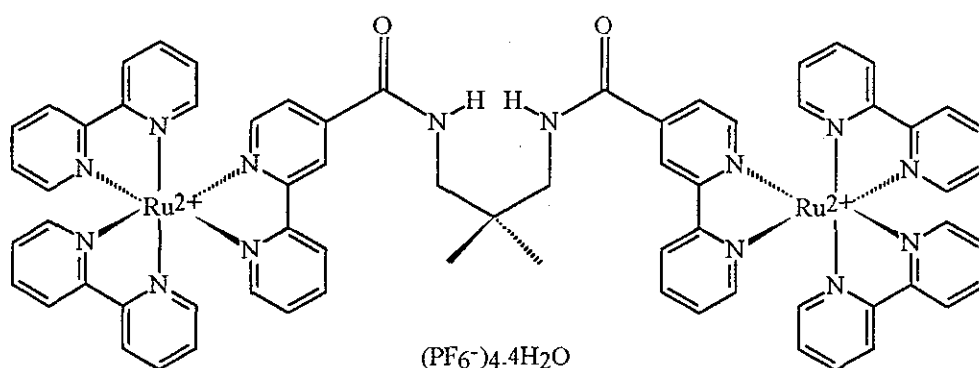
2



3



4



3.1 Anion complexation studies

3.1.1 ¹H NMR spectroscopy study

Extensive ¹H NMR spectroscopy titration investigations with Cl⁻ and H₂PO₄⁻ in acetonitrile were conducted by our collaborators at Oxford University to establish that the receptor, **1**, forms solution anion complexes of 1:1 stoichiometry. These anions were coordinated through a combination of attractive electrostatic and hydrogen bonding interactions with amide (CO-NH) and 3,3'-bipyridyl protons (Fig. 2).

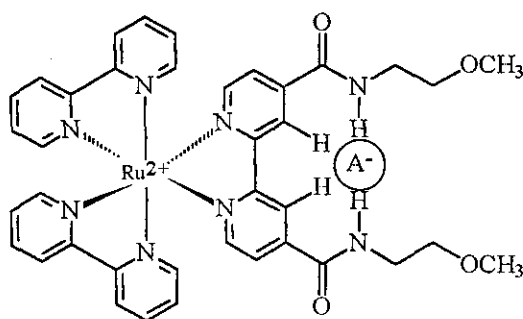


Fig. 2 Schematic diagram of halide binding by **1**, showing the hydrogen bonding interactions with amide (CO-NH) and 3,3'-bipyridyl protons.

Similar ^1H NMR titration experiments were performed with the complexes **2-5** by the Oxford research group. All of the complexes except for **3** showed significant proton shifts on the addition of Cl^- and H_2PO_4^- . This study showed that **2**, **4**, and **5** formed solution complexes with these anions. By contrast, **3** showed no significant proton shifts on addition of Cl^- , however, formation of a solution complex with H_2PO_4^- was established. With the ^1H NMR evidence in mind, anion recognition was probed using UV-vis spectroscopy, fluorescence emission spectroscopy and electrochemistry.

3.1.2 UV-vis and emission spectroscopy

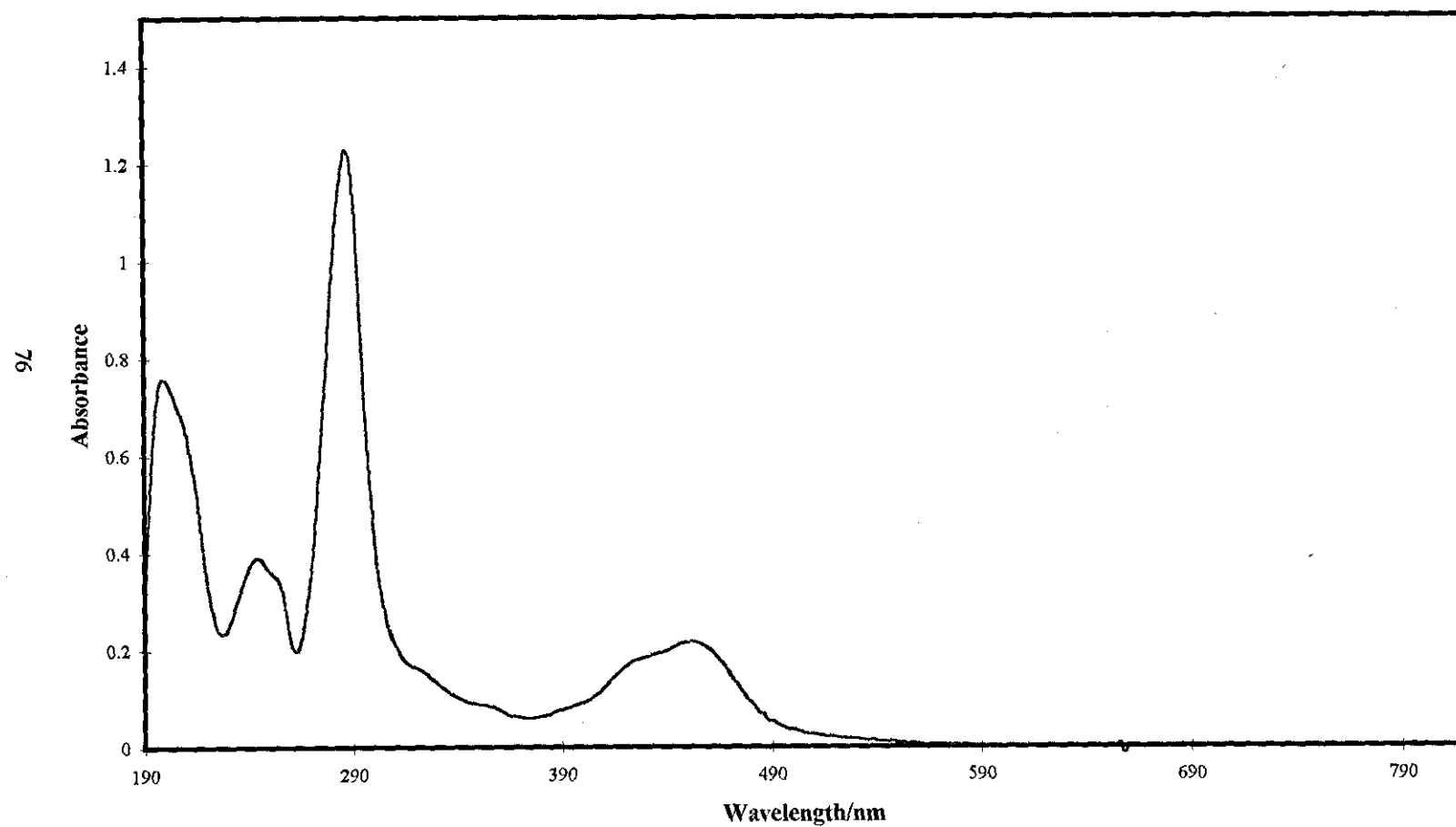
UV-vis spectra were recorded for all of the complexes in acetonitrile and the electronic absorbance data for $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, **1** and **2** is given in Table 1 and the absorbance spectrum of **1** is given in Fig. 3.

Table 1 Electronic absorption data in acetonitrile $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, **1** and **2**^a

$\lambda_{\text{max}}/\text{nm}$ ($10^{-4}\epsilon/\text{M}^{-1}\text{cm}^{-1}$)			
	$[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$	1	2
$\pi-\pi^*$ (2)	244 (3.23)	254 (4.82)	246 (3.52)
$\pi-\pi^*$ (1)	286 (9.82)	286 (9.63)	288 (7.82)
MC	352 (1.00)	350 (1.22)	352 (1.24)
MLCT	450 (1.74)	464 (1.54)	466 (1.84)

^a absorption band below 240 nm cannot be resolved from solvent. Solutions were 10^{-5} M.

Fig. 3 - Electronic absorption spectrum of $[\text{Ru}(\text{bipy})_3][\text{PF}_6]_2$ (1.25×10^{-5} M) in acetonitrile



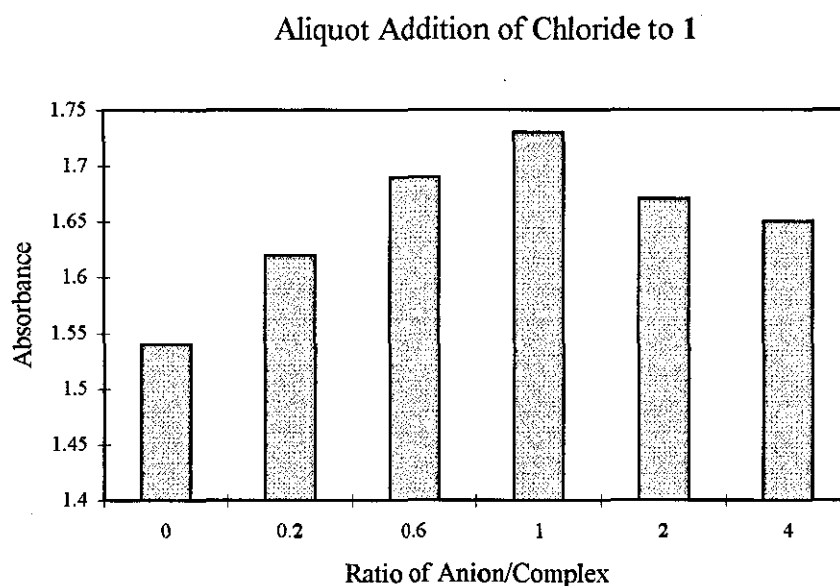
UV-vis spectroscopy was used to probe the binding of Cl^- on the aliquots addition of its tetrabutylammonium salt to $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, **1** and **2**. The metal-to-ligand charge transfer band (MLCT) (**Table 2**) was shown to be sensitive to the binding of Cl^- for **1** and indicated a 1:1 complex (**Chart 1**). As expected $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, and **2** showed little change on the addition of Cl^- . In the case of **2** the molecule has complexed its Cl^- counter anions such that there is no change in the absorption spectrum. Changes in the absorbance were noted on the addition of H_2PO_4^- to the solution of the complexes, but in the case of $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ a precipitate forms. The lack of change in the absorption spectrum of $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ on the addition of Cl^- and precipitation formation in the case of H_2PO_4^- adds further evidence to support anion binding by **1** and **2**.

Table 2 Electronic absorption data for the MLCT band in acetonitrile on the aliquot addition of chloride to $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, **1** and **2**^a

$\lambda_{\text{max}}/\text{nm}$ ($10^{-4}\epsilon/\text{M}^{-1}\text{cm}^{-1}$)			
equivalences	$[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$	1	2
0	450 (1.74)	464 (1.54)	466 (1.84)
0.2	450 (1.74)	464 (1.62)	466 (1.84)
0.6	450 (1.75)	464 (1.69)	466 (1.84)
1	450 (1.75)	464 (1.73)	466 (1.84)
2	450 (1.75)	464 (1.67)	466 (1.83)
4	450 (1.73)	464 (1.65)	466 (1.84)

^a Solutions were 10^{-5} M. Chloride was added as tetrabutylammonium salts to 4×10^{-4} M.

Chart 1



In the present instance, the most interesting band is that in the visible, assigned to the lowest energy singlet MLCT excited state. This band in the 4,4'-carbonyl amide disubstituted bipyridine ligand complexes **1-5** is red shifted (**Table 3**) compared with that of $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, because the electron-withdrawing carbonyl amide substituents increase the electron affinity of the ligands.

Table 3 Electronic absorption data for the MLCT band in acetonitrile for the complexes $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ and **1-5**^a

$\lambda_{\text{max}}/\text{nm} (10^{-4}\epsilon/\text{M}^{-1}\text{cm}^{-1})$					
$[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$	1	2	3	4	5
450 (1.74)	464 (1.54)	466 (1.84)	468 (1.50)	460 (1.11)	466 (3.04)

^a solutions were 10^{-5} M

Whilst changes in the absorbance spectra provided evidence for anion recognition, the effects were relatively small and the technique would be prone to interferences in an analytical application. Fluorescence emission spectroscopic measurements, which offer higher sensitivity and selectivity, were therefore carried out to probe anion binding. In real sample analyses, interference from other fluorescent species are likely to be negligible owing to the relatively high wavelength of the MLCT emission band in ruthenium(II) polypyridyl complexes.

As expected, the MLCT emission bands for **1-5** were red-shifted (**Table 4**) compared with that of $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ because of the lower energy of the MLCT absorbance as observed for the electronic absorption spectra.

Table 4 Fluorescence emission data in acetonitrile for $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ and **1-5**^a

$\lambda_{\text{max}}/\text{nm}$					
$[\text{Ru}(\text{bipy})_3][\text{PF}_6]_2$	1	2	3	4	5
594	623	620	628	607	627

^a Solutions were 10^{-6} M

On binding of Cl^- the fluorescence emission peaks for the complexes **1-3** showed blue shifts with significant intensity increases. These observations can be accounted for by considering the structure of the 4,4'-carbonyl amide disubstituted bipyridine ligand. In the uncomplexed forms of **1**, **2** and **3** the receptor sites on the bipyridyl group are free to rotate, and this lack of structural rigidity increases the chance of non-radiative decay.

On complexation, the Cl^- anion is bound by the amide receptor ligand. This restricts the rotation of the receptor sites which lead to the retrieval of the fluorescence which enhances the quantum yield. As predicted from NMR spectroscopy, no changes in the fluorescence emission spectrum of **4** took place on addition of Cl^- .

Complexes **1** and **4** were studied on the titrimetric additions of H_2PO_4^- . In the case of **1** the MLCT emission band on the binding of H_2PO_4^- showed a quenching in fluorescence (Fig. 4), which is contrary to the fluorescence enhancement on the addition of Cl^- . It was postulated that the structure of **1** is distorted on the binding of H_2PO_4^- , which leads to a decrease in quantum yield. As previously mentioned no change in fluorescence was observed on the addition of Cl^- to **4**. In contrast, the addition of H_2PO_4^- to **4** showed a substantial quenching in the fluorescence intensity (Fig. 5).

In the case of **4** it is envisaged that there will be significant repulsive interactions between the carbonyl oxygen lone pairs, which would result in inward pointing N-H groups. The N-H's in **4** are more acidic than in **1** due to electron withdrawal by pyridine on one side and by the carbonyl on the other. Inward pointing N-H's can bind the large H_2PO_4^- (suitably orientated oxygen lone pairs) whereas the bite angle of the N-H's is unsuitable to bind the smaller Cl^- (or possibly unsuitably oriented lone pairs).

As with the absorption measurement no changes were noted in the fluorescence spectra on addition of Cl^- and Br^- to $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, but on the addition of H_2PO_4^- a precipitate formed. These effects confirmed anion binding.

Fig. 4 - Fluorescence emission spectra of **1** (1×10^{-6} M) on the addition of H_2PO_4^- in acetonitrile

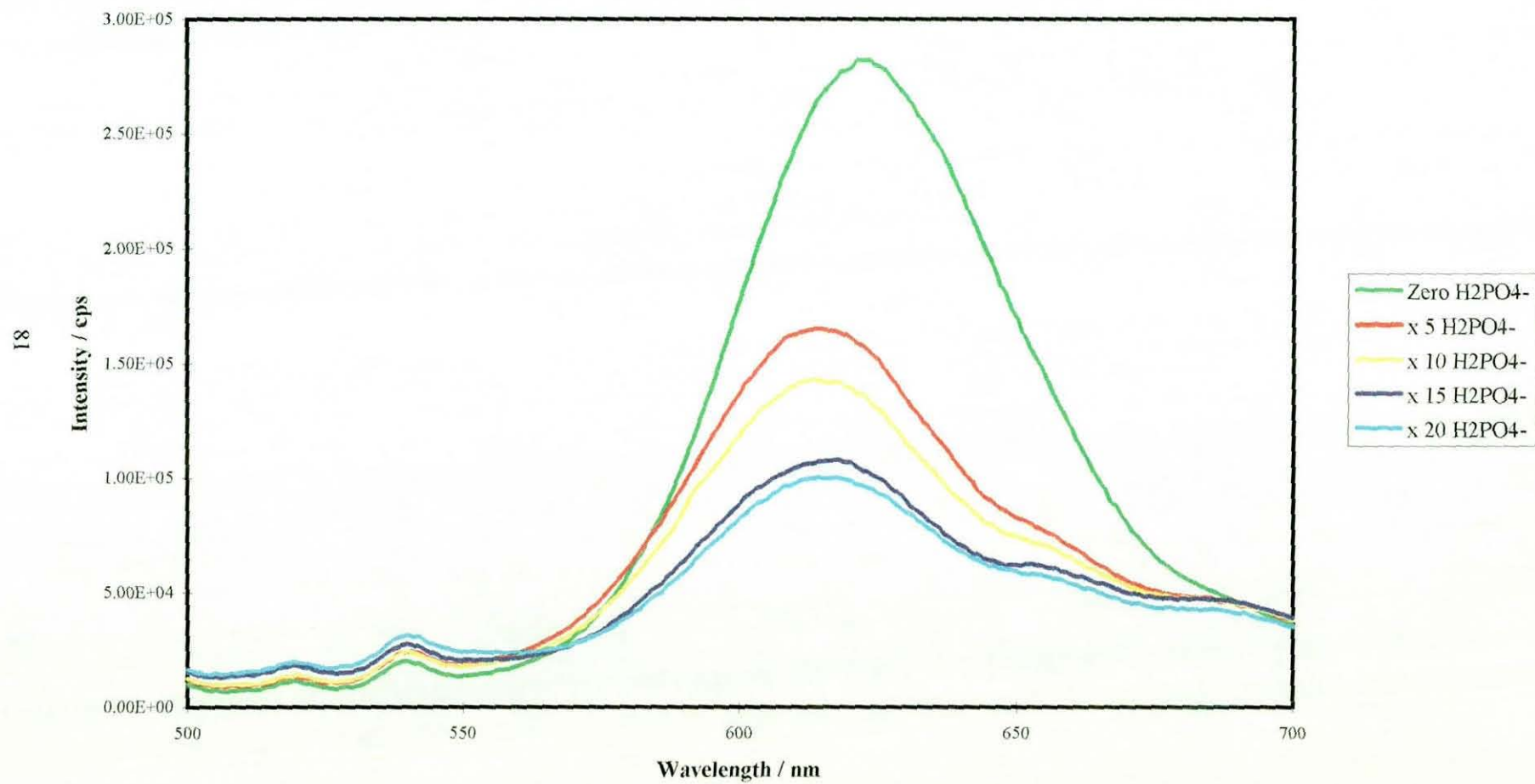
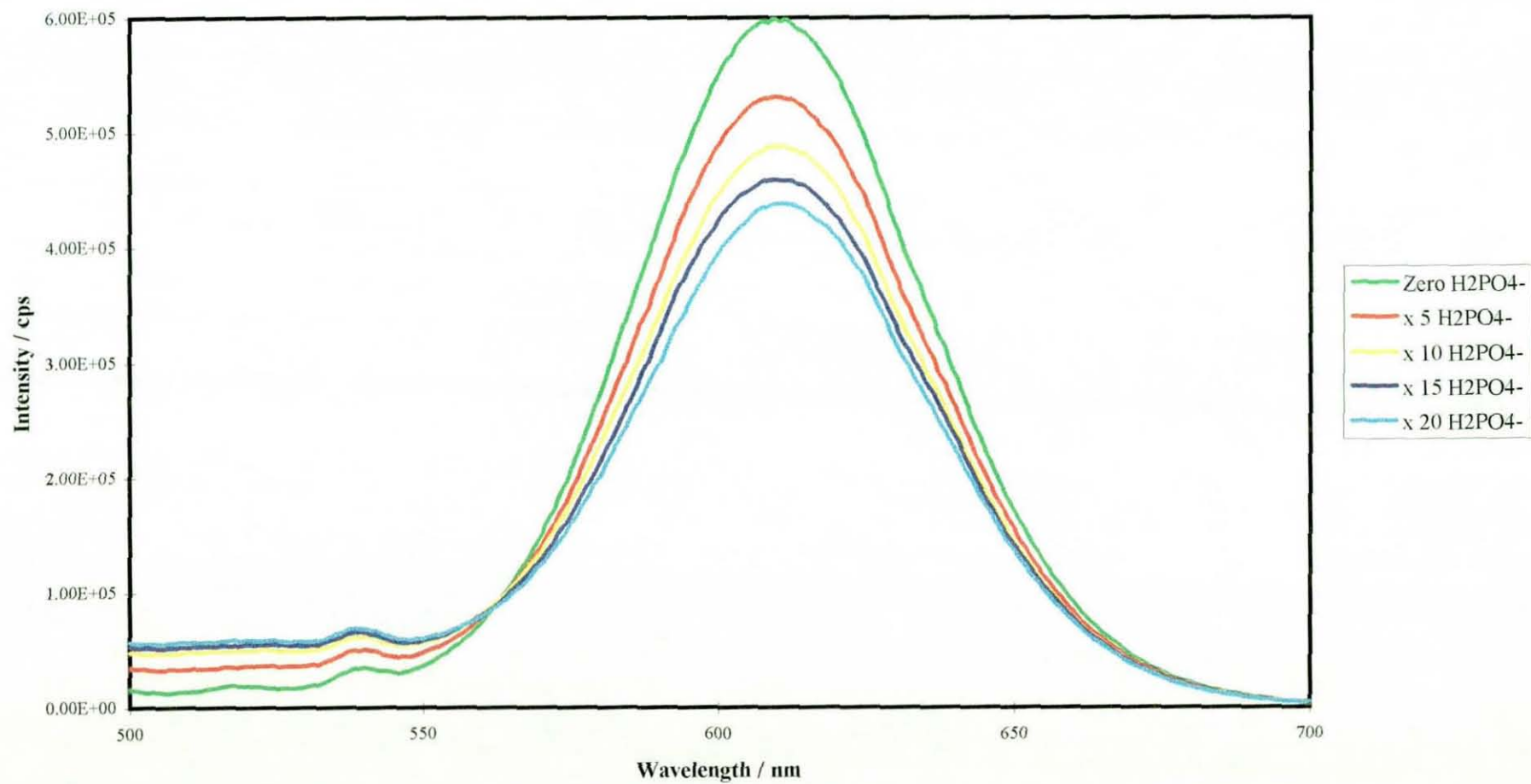


Fig. 5 - Fluorescence emission spectra of **4** (1×10^{-6} M) on the addition of H_2PO_4^- in acetonitrile



The titrimetric addition of various anions to **1** and **4** allowed the determination of the stability constants from fluorescence emission spectroscopy. **Tables 5** and **6** detail the stability constants and the effect on the fluorescence emission spectra. The stability constants were determined using the method by Valeur et al. which is the linear least squares analysis of plots of $I_f^0/(I_f^0 - I_f)$ versus the reciprocal of the anion concentration (**Formula 1 - see appendix 1 for derivation**).^{110, 111} The stability constant is given by the ratio of intercept/slope.

Formula 1

$$I_f^0/(I_f^0 - I_f) = [\epsilon_L \Phi_L / (\epsilon_{ML} \Phi_{ML} - \epsilon_L \Phi_L)] [1/K_s [X]] + 1]$$

I_f^0 = intensity of free ligand I_f = intensity of complexed ligand

Φ_L = quantum yield of the ligand ϵ_L = molar extinction coefficient of the ligand

K_s = stability constant ϵ_{ML} = molar extinction coefficient of the complex

$[X]$ = concentration of anions Φ_{ML} = quantum yield of the complex

Table 5 Fluorescence emission data for the titrimetric addition of various anions to **1** in acetonitrile.^a

Anion	Shift	Direction	Intensity	log K _s
Cl ⁻	5 nm	blue shift	enhanced	4.7
Br ⁻	3 nm	blue shift	enhanced	4.4
H ₂ PO ₄ ⁻	9 nm	blue shift	quenched	4.7

^a Solutions were 4x10⁻⁶ M and the stability constant (K_s) is represented as log K_s

Table 6 Titrimetric addition of various anions to **2** in acetonitrile^a

Anion	Shift	Direction	Intensity	log K _s
Cl ⁻	no change	no change	no change	no change
H ₂ PO ₄ ⁻	2nm	red shift	quenched	4.1

^a Solutions were 4x10⁻⁶ M and the stability constant (K_s) is represented as log K_s

From the stability constants it is possible to calculate the selectivity of a particular anionic guest for **1** (**Formula 2**).

Formula 2

$$\text{Selectivity} = \frac{\text{Stability constant for anion A}}{\text{Stability constant for anion B}}$$

Table 7 Selectivity of **1** with various anions

Anions	Selectivity
$\text{H}_2\text{PO}_4^-/\text{Cl}^-$	1.06
$\text{H}_2\text{PO}_4^-/\text{Br}^-$	2.18
Cl^-/Br^-	2.05

From the anion selectivity results for **1** in **Table 7** it can be seen that the ligand has very low discrimination ability between the various anions. This is probably due to the lack of structural rigidity of the acyclic ligand.

Complex **5** was shown to bind both H_2PO_4^- and Cl^- . The complex showed an enhancement in fluorescence on the addition of H_2PO_4^- and a quenching in fluorescence on the addition of Cl^- (**Table 8**). Aliquot addition of Cl^- anions to **5** allowed the determination of stability constants which was found to be $\log K_s = 4.5$. However the stability constant on the addition of H_2PO_4^- was not determined because at a 5 x excess the molecule is virtually fully complexed. This implies H_2PO_4^- forms a strong complex with **5** of an order $> \log K_s = 5$. The strength of the complex is probably due to the open structure which allows easy access to the binding sites for an anion such as dihydrogen phosphate. The λ_{max} also demonstrated a shift to higher energy of 9 nm on the addition of H_2PO_4^- whereas on the addition of Cl^- a shift of 3 nm to lower energy was observed. From these preliminary studies it is impossible to come to any conclusions as to the nature of the physical changes in intensity and λ_{max} shifts in **5**.

Table 8 Titrimetric addition of various anions to **5** in acetonitrile^a

Anion	Shift	Direction	Intensity	log K _s
Cl ⁻	3 nm	red shift	quenched	4.5
H ₂ PO ₄ ⁻	9 nm	blue shift	enhanced	>5

^a Solutions were 4×10^{-6} M and the stability constant (K_s) is represented as log K_s

The solvent environment was changed to 50%:50% (v/v) of acetonitrile (ACN) and dimethylsulphoxide (DMSO). DMSO (dielectric constant = 45) was chosen due to its higher dielectric constant than acetonitrile (dielectric constant = 37.5) which means that it is more likely to solvate the receptor cavity. It is proposed that this will lead to increased competitive binding between the more polar solvent and the anion. In this solvent environment, the addition of H₂PO₄⁻ to **1** (**Fig. 6**) causes a small increase in intensity and λ_{max} shift of 6 nm to higher energy. This suggests that the orientation of the bound H₂PO₄⁻ anion is changed from the pure acetonitrile case. For the 50%:50% (v/v) acetonitrile:DMSO solvent mixture, only slight intensity increases were found on addition of chloride (**Fig. 7**) or bromide (**Fig. 8**) at 100 x excess. This implies that the increased polarity of the solvent leads to the solvation of the guest cavity which resulted in competitive selectivity between the anions and DMSO.

3.1.3 Electrochemical recognition

As previously mentioned in the experimental (Chapter 2) the tris-2,2'-bipyridyl ruthenium(II) complexes undergo a metal-centred oxidation process and a series of

Fig. 6 - Fluorescence emission spectra of **1** (1×10^{-6} M) on the addition of H_2PO_4^- in acetonitrile/DMSO (50:50)

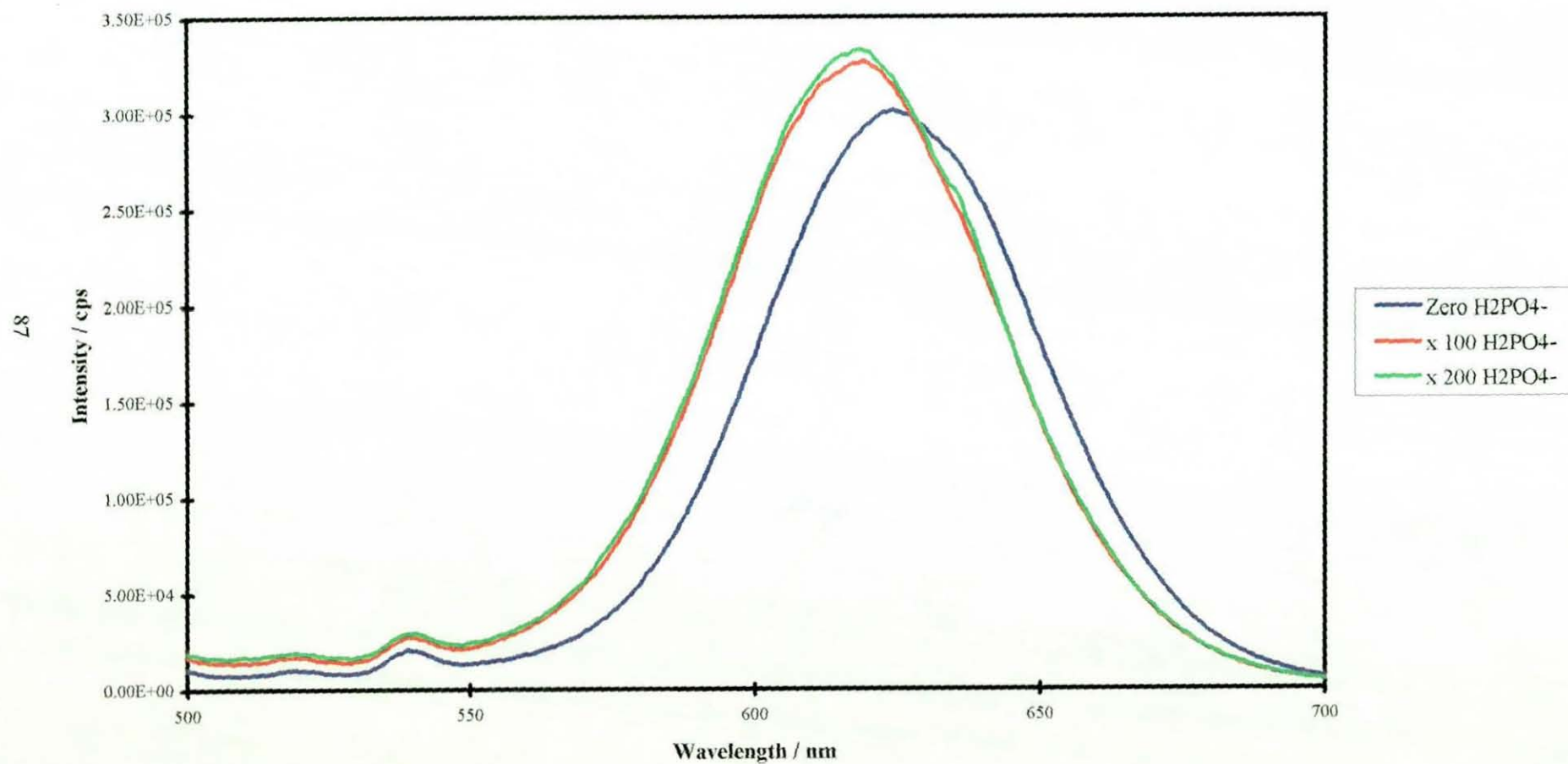


Fig. 7 - Fluorescence emission spectra of **1** (1×10^{-6} M) on the addition of Cl^- in acetonitrile/DMSO (50:50)

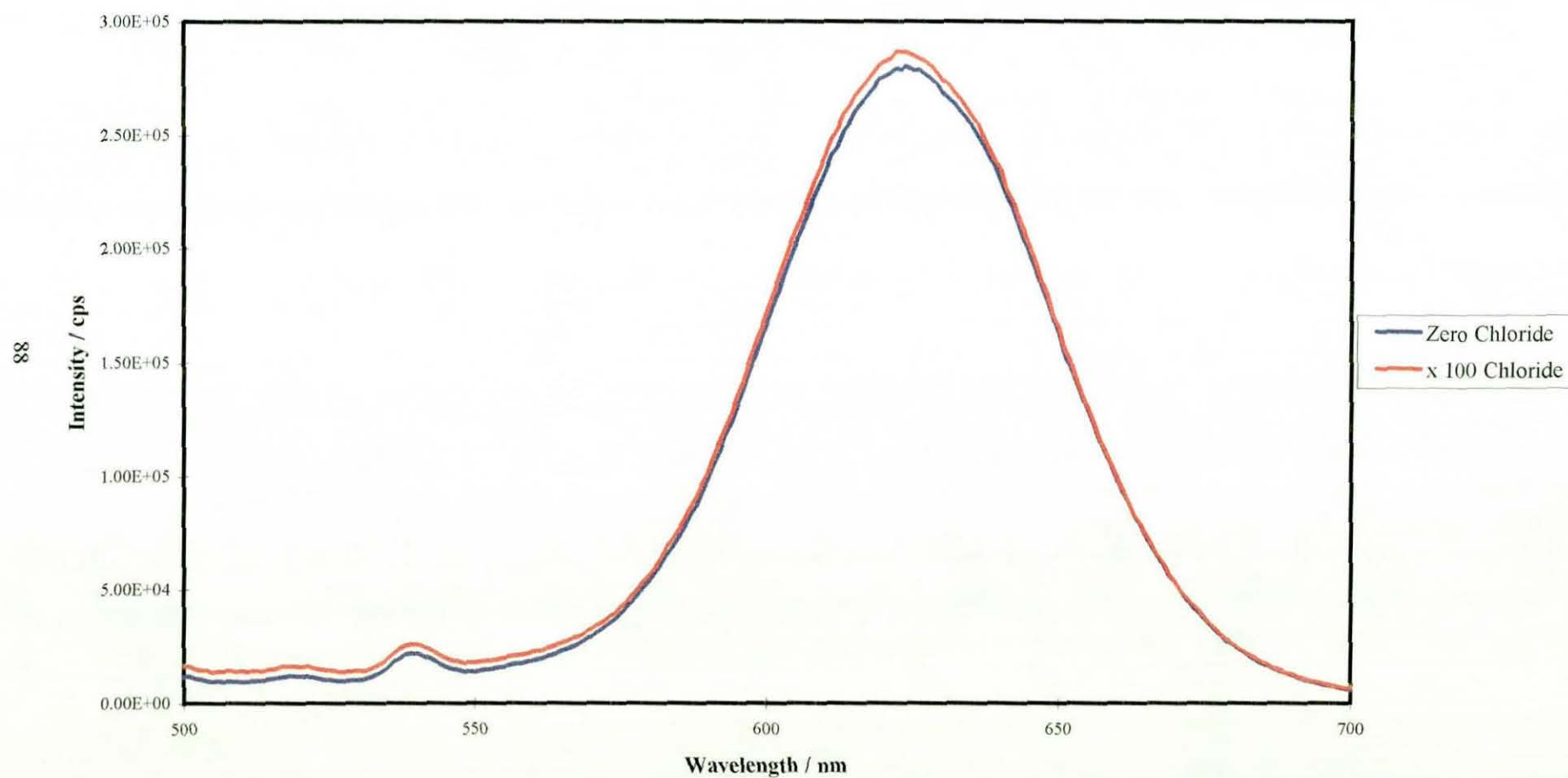
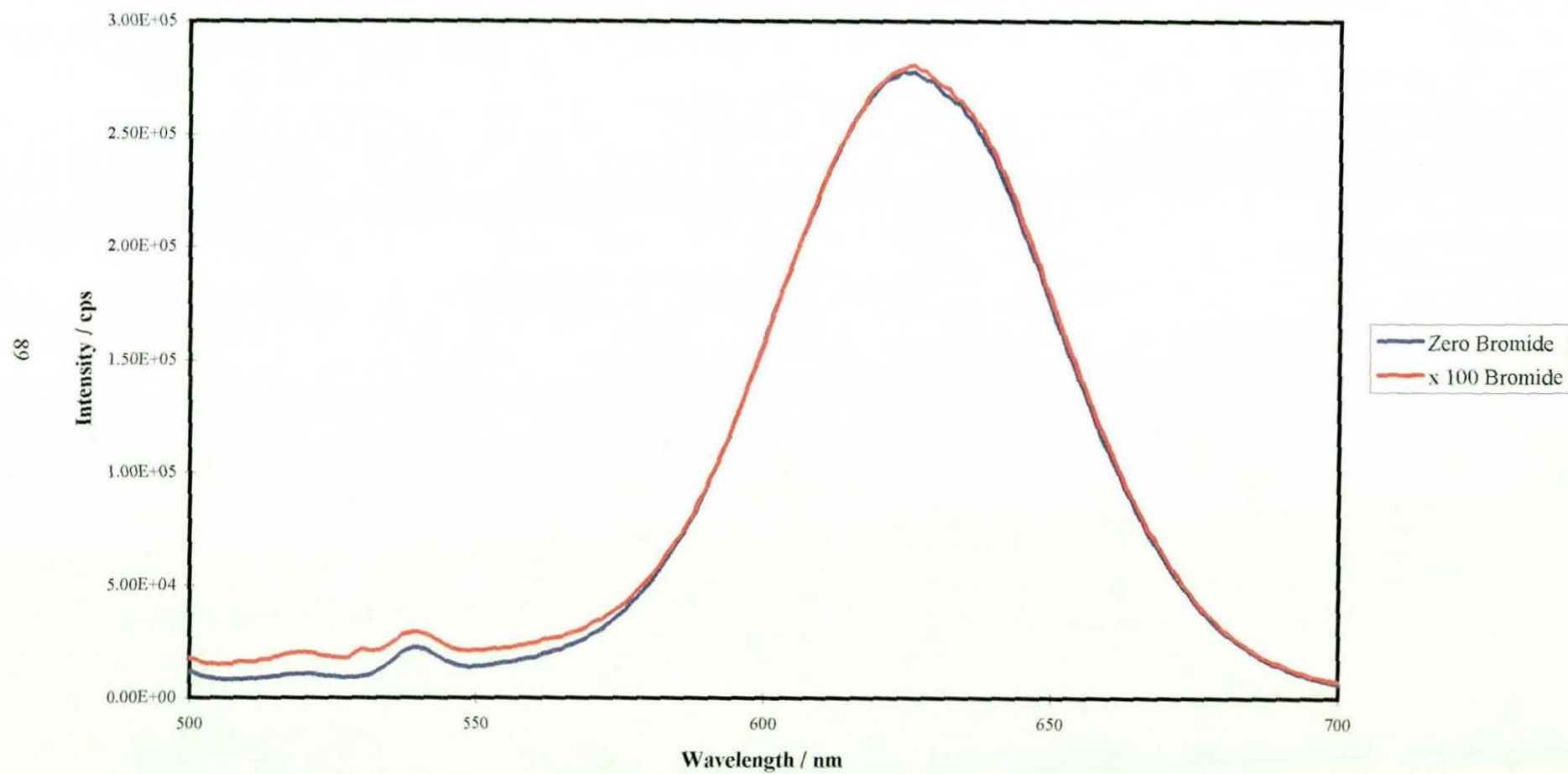


Fig. 8 - Fluorescence emission spectra of **1** (1×10^{-6} M) on the addition of Br^- in acetonitrile/DMSO (50:50)



three reduction waves corresponding to the successive one-electron reduction of the three ligands. Cyclic voltammetry was employed to investigate the electrochemical anion recognition properties of the complexes, redox potential data (**Table 9**) for the prototype $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ being in good agreement with the literature.¹⁰² The four redox potentials for **1** were anodically shifted compared with that of $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, owing to the electron-withdrawing nature of the carbonyl amide substituent. Where the shift for the least cathodic reduction was greatest, the redox wave can be assigned to the reduction of the carbonyl amide substituted ligand. It is this redox couple that exclusively undergoes a significant cathodic perturbation of 40mV on the addition of Cl^- anions (**CV 1**), suggesting an agreement with the ^1H NMR spectroscopy titration studies, that anion recognition takes place in the vicinity of the amide N-H protons. As expected from its lower polarising power, addition of bromide anions produced a smaller cathodic potential shift of 30 mV. No redox potential shifts were noted in analogous electrochemical anion recognition experiments with $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ which confirmed halide binding by the acyclic ligand.

CV 1 - CVs at 100 mVs^{-1} in an acetonitrile solution containing 0.2 mM of **1** in 0.1 M tetrabutylammonium tetrafluoroborate. **B** represents a 4 mM addition of tetrabutylammonium chloride to **A**. Applied potential versus saturated sodium chloride calomel reference electrode.

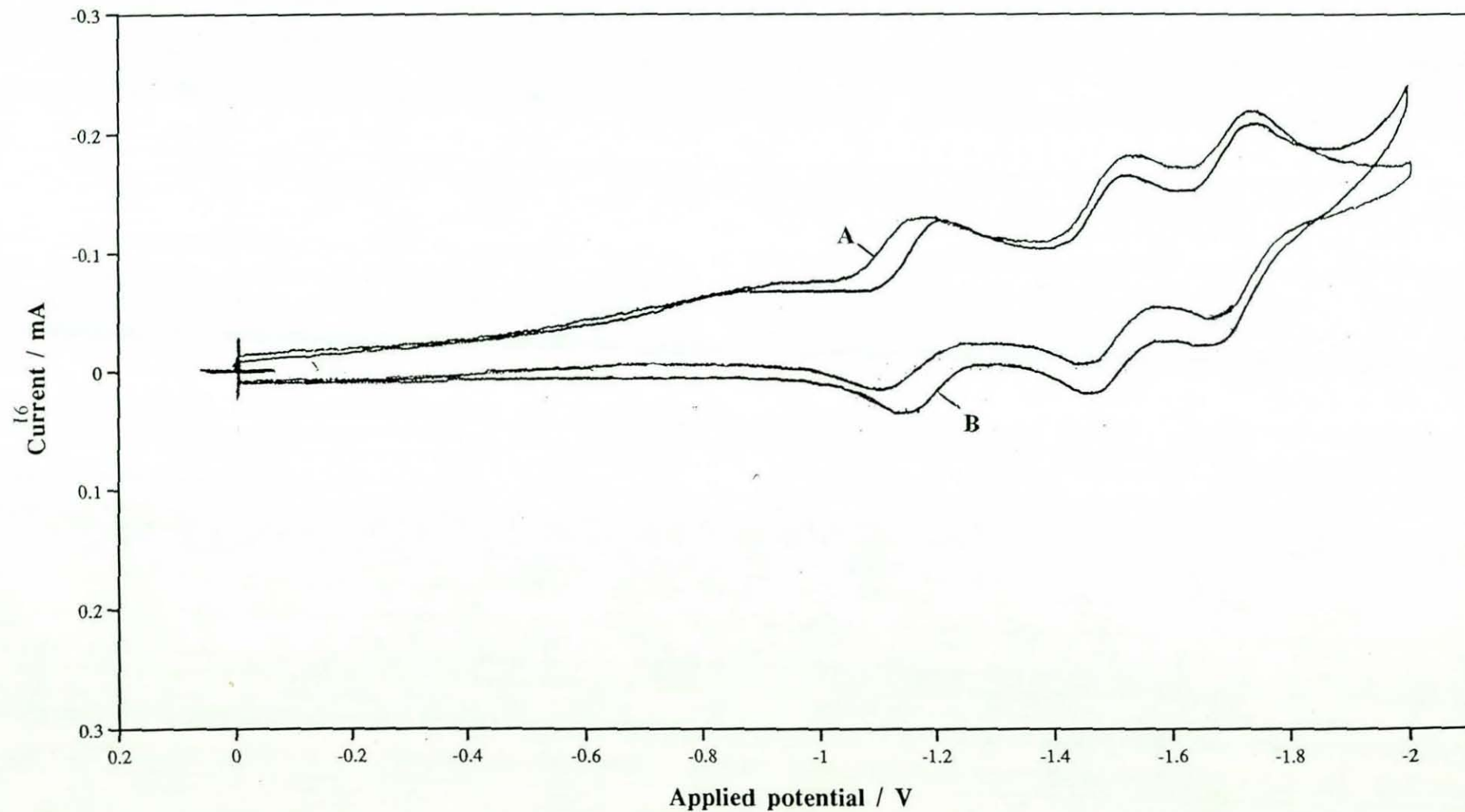
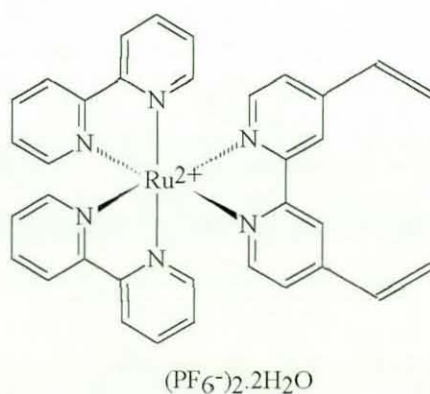


Table 9 Electrochemical data^a

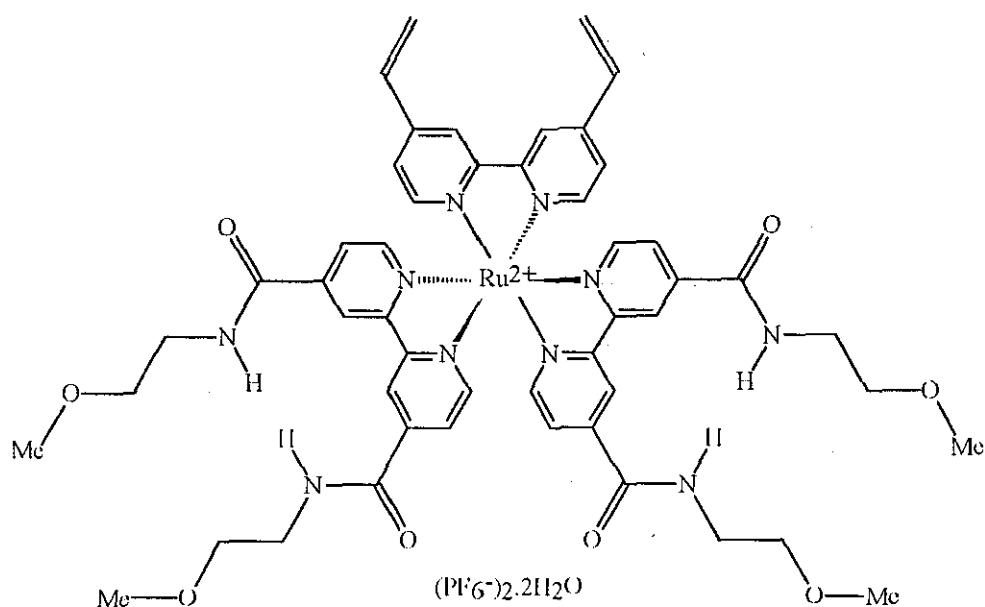
	metal based	bipyridyl reductions		
	$E^f(\text{Ru}^{2+/3+})$	$E^f(+2/+1)$	$E^f(+1/0)$	$E^f(0/-1)$
$[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$	1.33	-1.30	-1.50	-1.73
1	1.33	-1.12	-1.46	-1.68
1 + 20 equiv Br^-	1.33	-1.15	-1.46	-1.68
1 + 20 equiv Cl^-	1.33	-1.16	-1.46	-1.68

^a Obtained in acetonitrile solution containing 0.1 M Bu_4NBF_4 as supporting electrolyte. Solutions were 2×10^{-4} M in receptor, and potentials ($E^f = (E_p^A + E_p^C)/2$) were determined with reference to a saturated sodium chloride calomel electrode at 25 ± 2 °C at 100 mV s^{-1} scan rate. Cathodic shift perturbations of the first ligand-centred reduction couple produced by presence of Cl^- (20 equiv).

3.2 Polymer modified electrode studies



6



7

The ruthenium(II) trisbipyridyl derivative **7** incorporates two of the monoligands utilised in **1** which were shown to bind anions. More importantly the ruthenium(II) trisbipyridyl derivatives **6** and **7** were designed to contain two vinyl groups in order to produce a polymer modified electrode by reductive electropolymerisation. The technique utilised was the general reductive electropolymerisation method pioneered by Abruña et al.¹¹² It is envisaged that a polymer modified electrode made up of **7** would be capable of the electrochemical or spectrochemical detection of anions.

3.2.1 Electropolymerisation

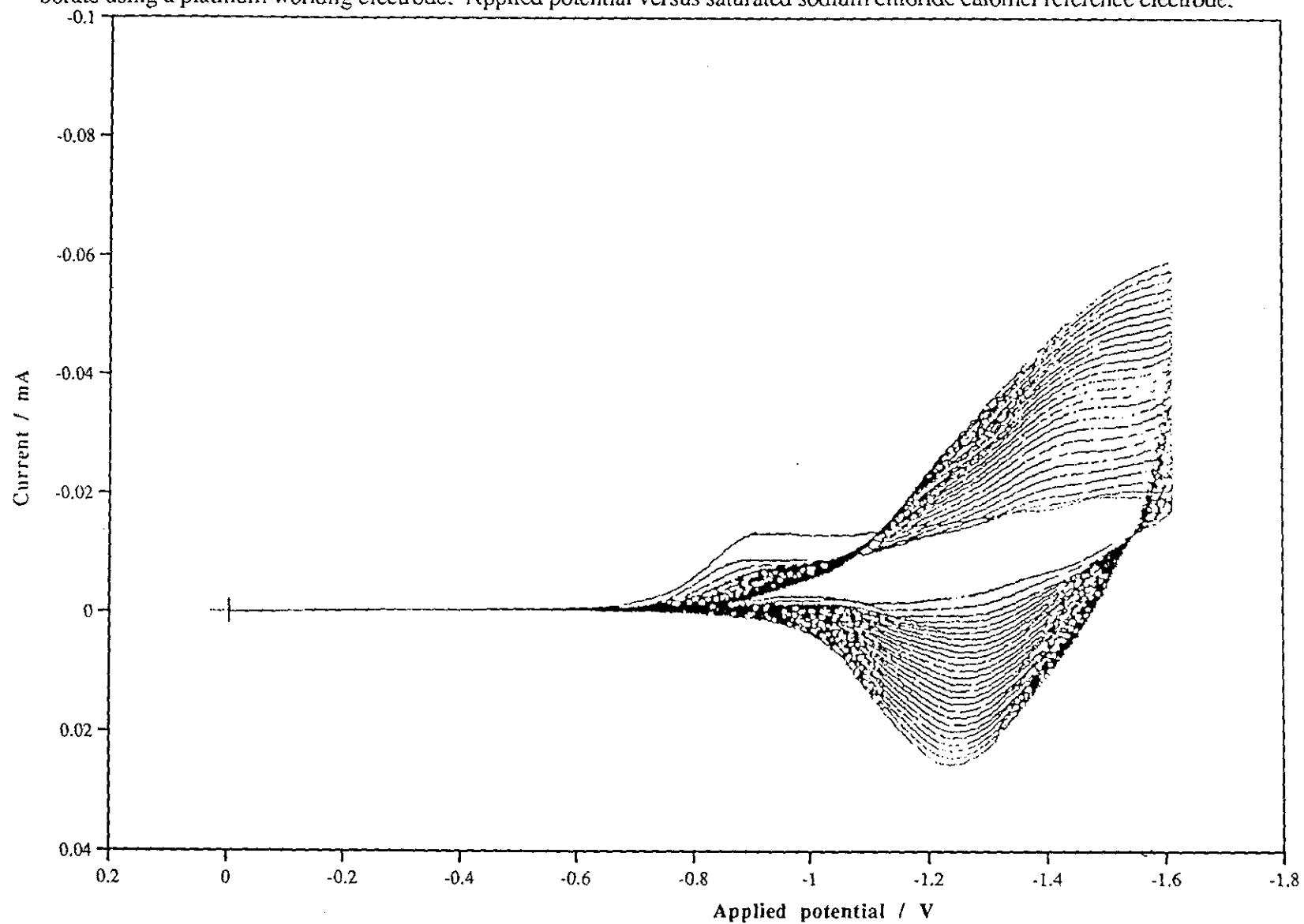
It was shown that **6** and **7** can be electropolymerised onto the surface of a platinum electrode and also onto an indium-tin oxide optically transparent conducting glass electrode (OTE). This was performed by sequential potential scanning of the monomer

solution from 0.0 V to -1.7 V at a scan rate (ν) of 100 mVs⁻¹ to the series of ligand-centred reductions, with the aim of activating the vinylic linkages and thus initiating electropolymerisation. The CVs 2 and 3 show the electropolymerisation of 7 on platinum and indium-tin oxide electrodes. Electropolymerisation occurred to form an adherent orange film on the electrode surface, this is exemplified by the cyclic voltammogram which shows a steady increase in current attributed to the combined electroactivity of the polymeric film and the inward-diffusing complex. Reductive electropolymerisation of Ru²⁺ complexes of vinyl substituted pyridyl ligands is thought to proceed via several different pathways, the most important of which is a radical-radical hydrodimerisation process involving pairs of vinyl groups.

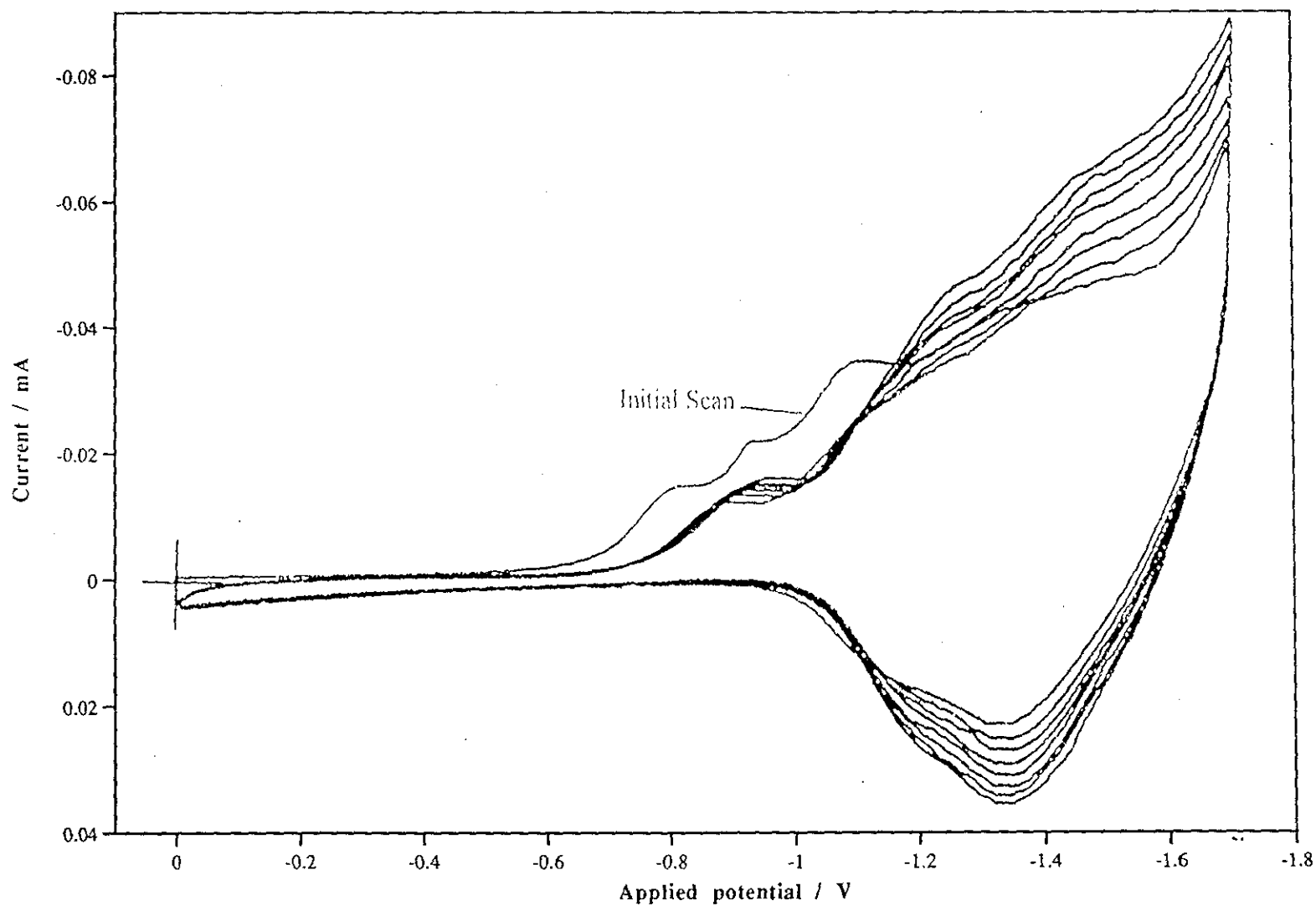
A stable metal-centred Ru^{2+/3+} redox process was observed for the electropolymerised 7 on the transfer of the polymer modified electrode to a complex-free solution. The dependence of peak currents (i_p) vs $\nu^{1/2}$ indicates diffusional control rather than surface control ($i_p \propto \nu$). High ΔE_p values which increase with scan rate suggest either slow electron-transfer kinetics or uncompensated Ohmic resistance in the polymer film. It was also noted that the polymer changed colour when held for a period of time at positive limits, this implies that all Ru²⁺ (orange) sites are oxidised to the Ru³⁺ (colourless). On oxidation to Ru³⁺ the loss of the MLCT band was observed which resulted in the colourless polymer film.

On the initial scan an irreversible oxidation of the metal centre occurred (CV 4). The true nature of this oxidation peak is not known but it is thought to be a sort of "breaking in" of the polymer film. Murray et al. proposed that the "breaking in" of the polymer

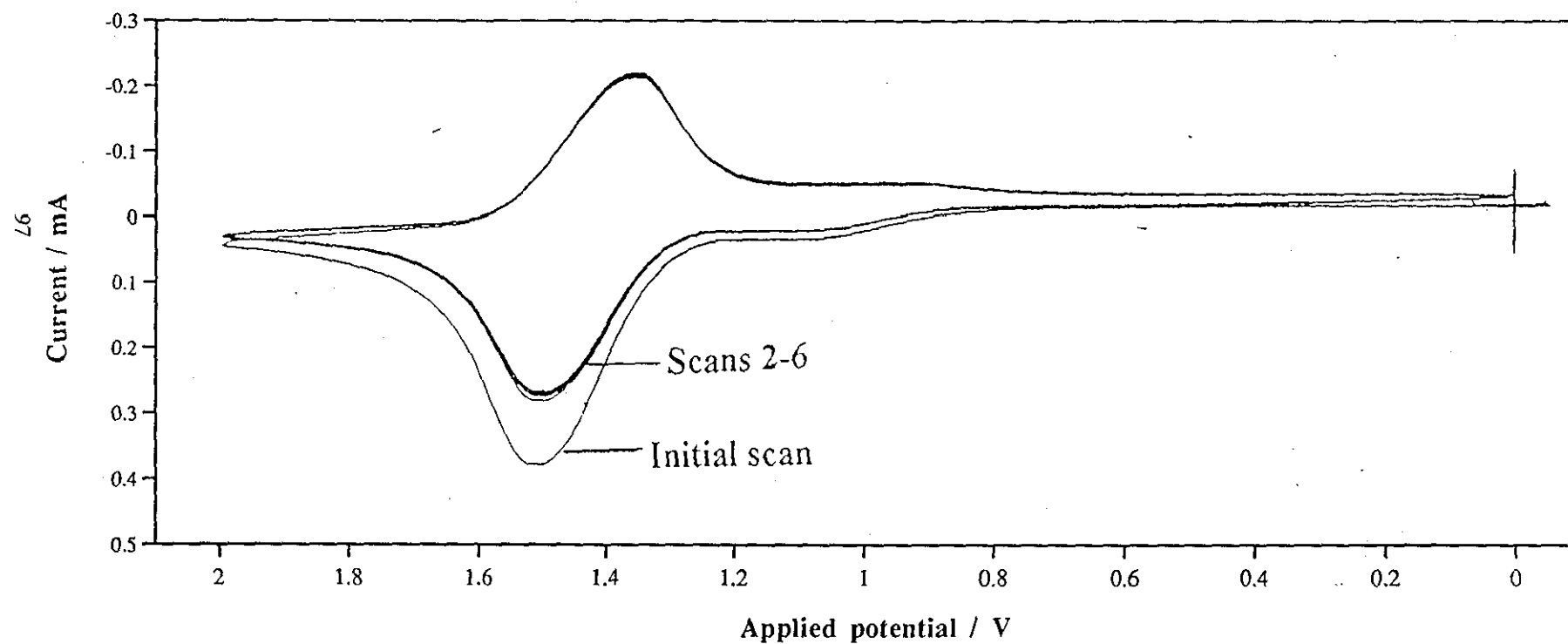
CV2 :- Sequential scans at 100mVs^{-1} in an acetonitrile solution containing 0.2mM of 7 in 0.1 M tetrabutylammonium tetrafluoroborate using a platinum working electrode. Applied potential versus saturated sodium chloride calomel reference electrode.



CV3 :- Sequential scans at 100mVs^{-1} in an acetonitrile solution containing 0.2mM of **7** in 0.1 M tetrabutylammonium tetrafluoroborate using an indium-tin oxide working electrode. Applied potential versus saturated sodium chloride calomel reference electrode.



CV 4 - Sequential CVs at 100 mVs^{-1} of a polymer modified electrode (indium-tin oxide) of 7 in an acetonitrile solution containing 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride reference electrode.



film is due to a non-uniform film environment where some of the Ru^{3+} sites are isolated and are not reduced back to Ru^{2+} . On holding the polymer at 0.0 V the "breaking in" is observed again, which implies that all the sites are returned to the Ru^{2+} state.

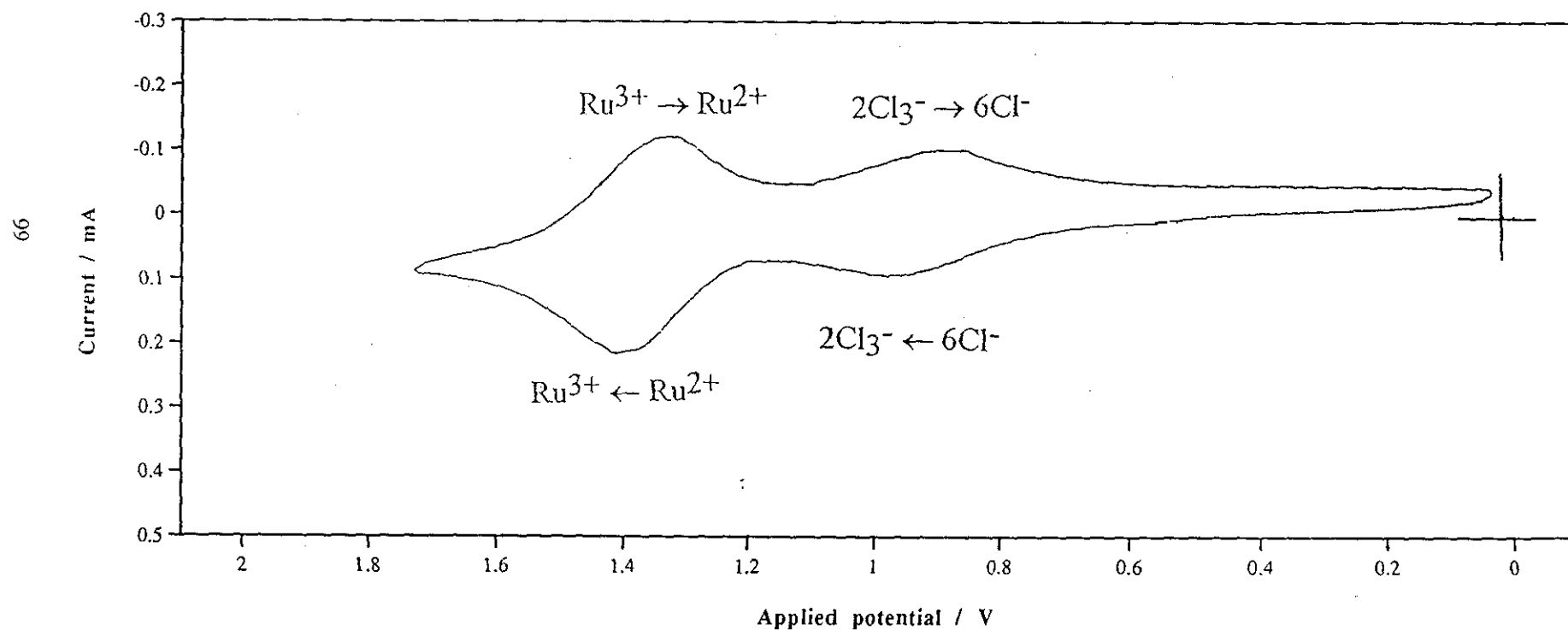
It was considered that the ligand centred redox processes would, however, be sensitive to anion binding. Unfortunately, it was not possible to monitor the ligand centred redox processes due to the broad nature of the vinyl-substituted ligands redox processes in the polymer-modified electrode.

As expected no perturbation of the metal-centred $\text{Ru}^{2+/3+}$ redox process was noted on the addition of Cl^- anion to the polymer films **6** (CV **5**) and **7**. This was not surprising as $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ and **1** showed no perturbation in the metal-centred $\text{Ru}^{2+/3+}$ redox process on the addition of anions. However the redox process associated with Cl^- can be observed on the addition to the polymer films of **6** and **7**. In the previous cases the receptor sites of **7** have been shown to complex anions therefore the redox process of the Cl^- anion was expected. However, complex **6** also showed the take up of Cl^- anion, this is not that surprising as the Cl^- will be taken up as a counter anion to the Ru^{2+} metal centre.

3.2.2 Emission spectroscopy

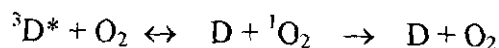
Spectrochemistry has proved to be a superior technique to electrochemistry in the study of the complexation of anions in the monomer form (section 3.1.2). Therefore it was a natural progression to study the spectrochemistry of the polymer film. A freshly

CV5 :- CV of a polymer modified electrode (6), on the addition of Cl^- , after its transfer to a clean supporting electrolyte of 0.1 M tetrabutylammonium tetrafluoroborate in acetonitrile.



prepared polymer coated OTE was produced for fluorescence measurements. Fluorescence studies were conducted by positioning the polymer coated electrode in the cell holder at around 45° to the excitation beam. Care was taken to prevent reflectance of the excitation beam to the detector. No fluorescence was noted for any of the samples when the measurements were carried out in air. As with other [Ru(bipy)₃(PF₆)₂] complexes, the quenching of fluorescence may occur due to charge transfer (CT) interaction with oxygen (Scheme 1).¹¹³

Scheme 1 Quenching reaction of the excited state by oxygen



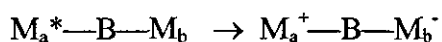
D represents the [Ru(bipy)₃(PF₆)₂] derivative

³D* represents the triplet excited state of the [Ru(bipy)₃(PF₆)₂] derivative.

In order to prevent the interaction of oxygen with the polymer film the sample was placed in acetonitrile and purged with nitrogen. Fluorescence was observed from the sample, however on removal of the sample from solution fluorescence was still observed. This was interpreted as being due to trapped monomer leaching out of the polymer film into solution. The sample was repeatedly washed with acetonitrile to removal all the trapped monomer until no fluorescence was observed. It was postulated that the fluorescence from the polymer film could be quenched by a variety or combination of routes (Scheme 2).¹¹⁴

Scheme 2. Possible quenching routes

- (i) Quenching by the indium-tin oxide and platinum electrodes
- (ii) Quenching due to the electrochemical supporting electrolyte in the film
- (iii) Photo induced electron transfer

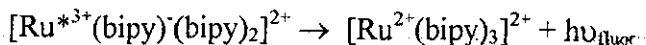
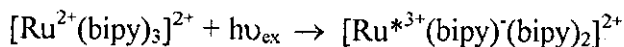


M_a and M_b represents two metal-containing fragments and B is a bridging ligand.

One of the possible quenching mechanisms may be due to the electrode materials which may quench the excited state of the polymer films of 6 and 7. This was disproved by sonicating the polymer film off the electrode surface into solution and still no fluorescence was observed. If the presence of the supporting electrolyte was the source of the quenching then repeated washing of the electrode should yield fluorescence, this is not the case. Which leads to quenching due to photo induced electron transfer. Fluorescence occurs due to the metal-to-ligand charge transfer band (MLCT). On excitation of the MLCT band the metal centre (Ru^{2+}) is oxidised to the excited (Ru^{3+}) resulting in the transfer of an electron to the ligand (Scheme 3). The monomer soon loses (μs timescale) the energy of excitation by non-radiative decay and more importantly by radiative decay (fluorescence). For the polymer this is further complicated by the linking of the metal-centres which facilitates the transfer of an electron which can deactivated the excited metal-centre (preventing fluorescence) (Scheme 4).

Scheme 3 Excitation of the photo-active centre

Monomer



Scheme 4

Polymer

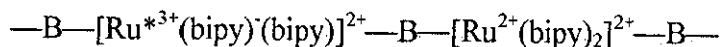
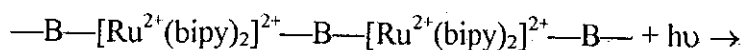
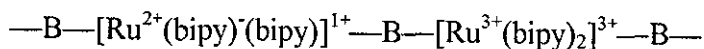
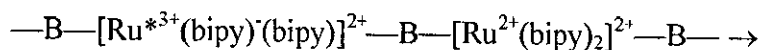


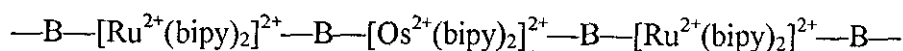
Photo-induced electron transfer - quenches the excited state due to electron transfer



B is the bridging ligand - the product of the electropolymerisation of dvbipy

The quenching of the fluorescence via electron transfer for the polymeric ruthenium(II) trisbipyridyl derivatives prevent the use of the molecule in the development of a spectrochemical sensor. Photoinduced electron transfer would probably be prevented if the polymer film was built up of mixed metal centres. This is because the $[\text{Os}(\text{bipy})_3]^{2+}$ moiety has a lower oxidation potential than the $[\text{Ru}(\text{bipy})_3]^{2+}$ (Scheme 5). This should isolate the $[\text{Ru}(\text{bipy})_3]^{2+}$ metal centres leading to the observation of fluorescence.¹¹⁴

Scheme 5 Mixed metal centres



B is the bridging ligand - the product of the electropolymerisation of dvbipy

3.3 Polymer modified electrodes as anion filters

A possible use of the electropolymerisable ruthenium(II) trisbipyridyl derivative **7** is to produce filtration systems for commercial important or environmental hazardous anions. An example of such an environmental important anion is pertechnetate (TcO_4^-) which is very similar in structure to that of dihydrogen phosphate (H_2PO_4^-) anion.^{115, 116}

Technetium (Tc) is produced as a by-product from the nuclear fuel cycle (40 kg of ^{99}Tc per reactor each year).

Important reasons for the removal of Technetium :-

1. It can be used as a radio nuclide, in nuclear medicine
2. TcO_4^- is extracted as the counter ion to UO_2^+ (Purex Process)
3. $\text{UO}_2(\text{NO}_3)_2(\text{TBP})_2 + \text{TcO}_4^- \leftrightarrow \text{UO}_2(\text{NO}_3)(\text{TcO}_4)(\text{TBP})_2 + \text{NO}_3^-$
4. Poor separation means that Tc goes into the highly radioactive waste (HAW)
5. Re-use of U from the nuclear fuel cycle may also cause problems if it contains measurable amounts of Tc: Fluorination to UF_6 for isotope separation leads to the formation of TcF_6 which, due to its high volatility, is easily discharged into the air.

A number of reports state that there is great interest in the:

- quantitative transfer of Tc into one waste stream
- development of selective separations of Tc.

The anion binder 7 has been successfully electropolymerised onto a commercially available steel stainless filter. In principle if the stainless steel filter was incorporated into a contaminated stream this should provide a method for the removal of environmental/commercial important anions.

The filter provides a honeycomb of paths which can be penetrated by a solution of 7 this should lead to the formation of polymer coating deep into the internal structure. Which results in electropolymerisation of the solution to yield a polymer coated filter with a large surface area.

An on-line filtration system such as this may result in the removal of pertechnetate from the aqueous stream. However the effect of water on complexation and denaturation of the polymer film due to radiation also needs to be quantified. Radiolysis by ^{99}Tc which is a very low energy β^- emitter and has a half-life of 2.13×10^5 years is expected to be very small. Unfortunately the facilities available have made it impossible to study this further.

3.4 Conclusions

The Lewis acidic redox-active and photo-active ruthenium(II) trisbipyridyl moiety in combination with amide CO-NH groups has been successfully incorporated into acyclic structural frameworks to produce novel receptors capable of the electrochemical and spectral recognition of anions. Electronic absorption and emission spectroscopy measurements on the complexes **1**, **2**, **3** and **5** have shown that these receptors are sensitive to the binding of anions such as Cl^- and H_2PO_4^- . Emission spectroscopy has proved to be the most useful tool in probing the interactions of the receptors and anions in acetonitrile. Significant perturbations in the MLCT λ_{max} and quantum yields were observed in the receptors **1**, **3**, and **5**. More importantly exclusive selectivity to the binding of H_2PO_4^- anion in preference to Cl^- by receptor **4**. The solvent environment was changed from acetonitrile to a 1:1 (v/v) mix of acetonitrile and DMSO. Changes in the emission spectra of **1** were small on the aliquot addition of Cl^- and Br^- with H_2PO_4^- showing the largest changes, which is thought to be due to competitive solvation of the cavity. This was expected as the acyclic structure could readily be opened up by a polar solvent such as DMSO preventing the binding of anions.

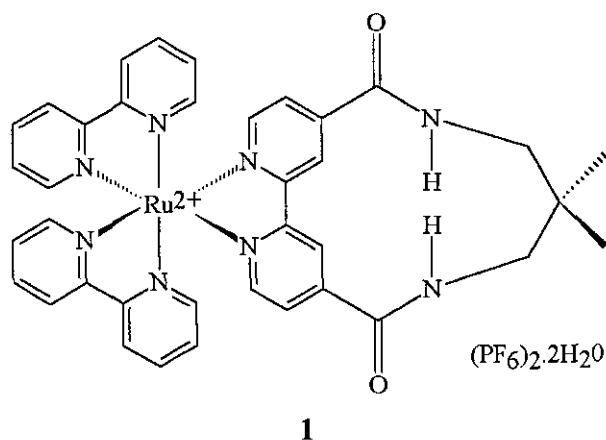
Electrochemical investigations have demonstrated that receptor **1** electrochemically recognises Cl^- and Br^- . These novel acyclic receptors offer the dual capability of detecting and sensing anions via electrochemical and optical methodologies.

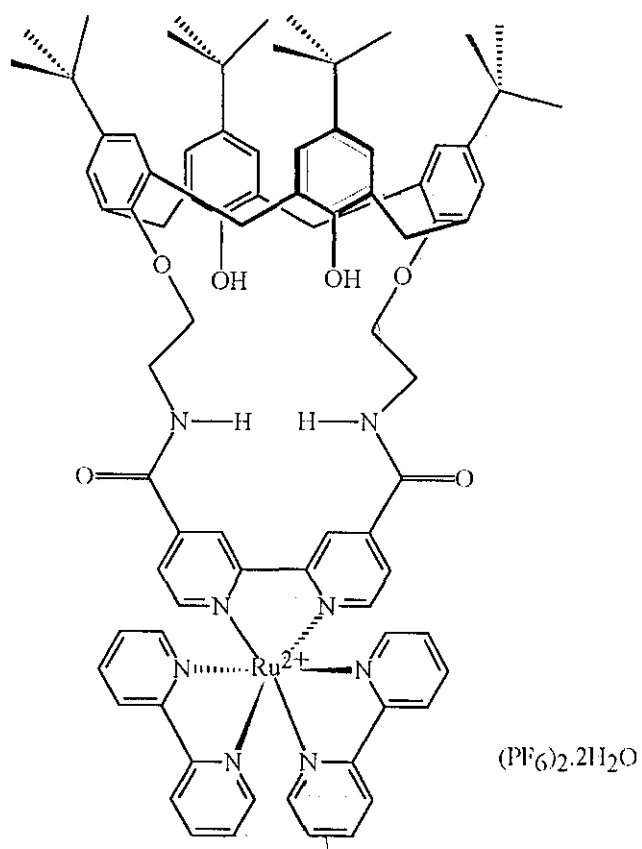
The complexes **6** and **7** were successfully reductively electropolymerised to produce a polymer modified electrode. It was hoped that in the case of **7** incorporating CO-NH anions sites that electrochemistry and spectrochemistry could be used in the probing of anion binding. However, photo-induced electron transfer prevented the use of emission spectroscopy and in the case of electrochemistry the masking effects of the vinylic redox processes prevented the study of anion recognition using the anion sensitive ligand. On examination of the metal-centred redox processes of **6** and **7** no perturbation was observed on the addition of chloride. Redox processes associated with the uptake of chloride by the polymer modified electrode were observed, which lead to the proposal that the polymer could be used in the extraction of commercially or environmentally important anions.

Chapter 4 -Macrocyclic and Calix[4]arene Ruthenium(II) Bipyridyl Receptor Molecules

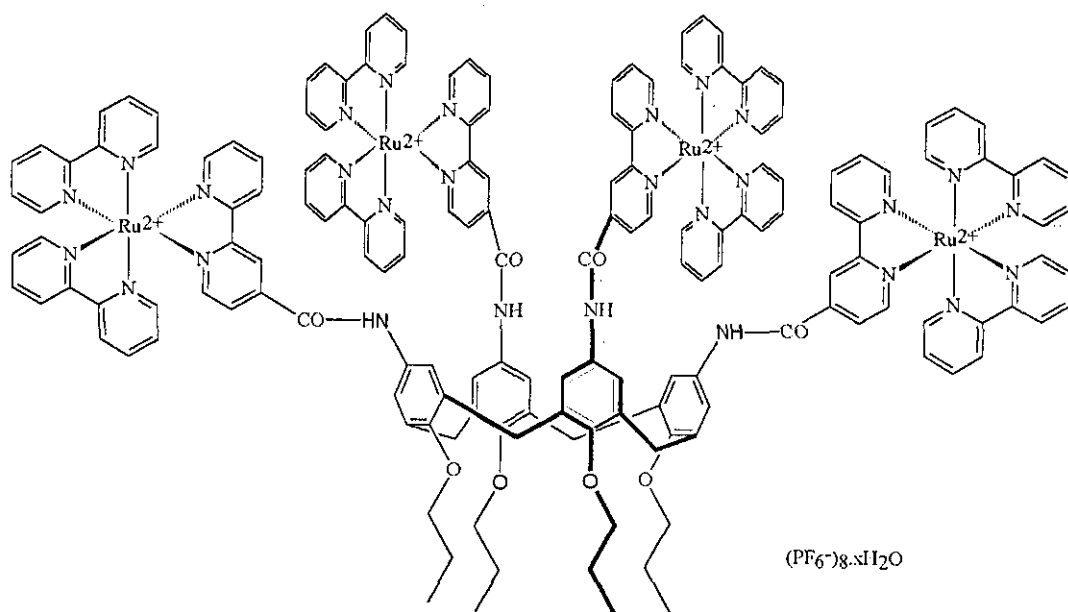
4.0 Introduction

The previous chapter demonstrated that suitably designed acyclic receptors bind anions. However acyclic receptor molecules have very low structural rigidity which leads to poor selectivity. It was therefore envisaged that the incorporation of macrocyclic and calix[4]arene moieties into receptors **1-3** would greatly enhance the rigidity of the molecule and thus increase selectivity.¹⁰⁷ Increased structural rigidity in the receptor site makes it possible to preorganise the binding sites, such as designing a tetrahedral binding site for a tetrahedral guest molecule. Guest selectivity can also be altered due to size exclusion with the formation of a cavity of predictable size.





2



3

4.1 Spectroscopic anion complexation studies

4.1.1 ^1H NMR spectroscopy study

The recognition of anions in solution was initially studied by ^1H NMR spectroscopy by Beer et al.¹⁰⁷ They found that additions of tetra-butyl ammonium chloride or dihydrogen phosphate to DMSO- d_6 solutions of **1**, resulted in remarkable perturbations of the respective receptor's protons. For example, with Cl^- the amide protons of **1** shifted downfield by $\Delta\delta$ 1.2 ppm after the addition of one equivalent of anion. Also the 3,3'-bipyridyl protons of **1** are substantially perturbed. They also noted that receptor **1** exhibited a large magnitude of selection between H_2PO_4^- over Cl^- . Analogous ^1H NMR spectroscopy titration experiments were carried out with the calix[4]arene containing receptor **2** with tetra-butyl ammonium chloride and dihydrogenphosphate salts in DMSO- d_6 . The selectivity trend $\text{H}_2\text{PO}_4^- > \text{Cl}^-$ is shown by receptor **2**. Beer et al. also noted for H_2PO_4^- that the stability constant showed an increase in magnitude for the acyclic alkyl receptor (**1** - Chapter 3) < macrocyclic receptor **1** < calix[4]arene receptor **2**.¹⁰⁷ This supports the idea that the rigidity increases the levels of selectivity and stability of the resulting complex. Preliminary ^1H NMR spectroscopy studies have also been carried out on **3**, which shows that it binds both Cl^- and H_2PO_4^- .

4.1.2 Electronic absorption and emission spectroscopy

Ruthenium(II) trisbipyridyl complexes exhibit metal-centred (MC), intraligand ($\pi-\pi^*$) and low energy metal-to-ligand charge transfer (MLCT) absorption bands. The

experiments were performed in the more polar DMSO rather than acetonitrile due to the increased rigidity of the receptor site. Increased rigidity of the receptor site prevents extensive distortion. Electronic absorption λ_{max} and ϵ data for **1**, **2** and **3** in DMSO are summarised and contrasted with those of the prototype $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ in **Table 1**. The most important band is in the visible region, assigned to the lowest energy singlet MLCT excited state. As with the acyclic ruthenium(II) trisbipyridyl receptor (Chapter 3) the MLCT band of the complexes **1-3** are red shifted compared with that of $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, because the electron-withdrawing carbonyl amide substituents increase the electron affinity of the ligands.

Table 1 Electronic absorption data in DMSO for $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, **1**, **2**, and **3**^a

$\lambda_{\text{max}}/\text{nm} (10^{-4}\epsilon/\text{M}^{-1} \text{ cm}^{-1})$				
	$[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$	1	2	3
$\pi-\pi^*$ (1)	292 (9.87)	292 (10.50)	292 (12.01)	292 (31.46)
MC	356 (0.39)	358 (1.40)	354 (1.74)	*
MLCT	456 (1.92)	470 (2.45)	466 (2.44)	460 (8.07)

^a absorption bands below 240 nm cannot be resolved from solvent. Solution concentrations for $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, **1**, **2** were 10^{-5} M and 4×10^{-6} M for **3**

* not resolved

UV-vis spectroscopy was used to probe anion binding on the addition of Cl^- and H_2PO_4^- to the complex $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, **1**, **2** and **3**. The lack of changes in the

absorbance of $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ on the addition of Cl^- and precipitate formation for H_2PO_4^- indicates that this complex is insensitive to the binding of anions (**Table 2**).

Table 2 Electronic absorption data in DMSO on the addition of Cl^- and H_2PO_4^- to $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]^a$

$\lambda_{\text{max}}/\text{nm}$ ($10^{-4}\epsilon/\text{M}^{-1}\text{cm}^{-1}$)			
$[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$	Zero addition	Cl^-	H_2PO_4^-
$\pi-\pi^*$ (1)	292 (9.87)	292 (10.9)	ppt
MC	356 (0.39)	356 (0.39)	ppt
MLCT	456 (1.92)	456 (1.95)	ppt

^a Absorption bands below 240 nm cannot be resolved from solvent. Solutions were 10^{-5} M. Cl^- and H_2PO_4^- were added as tetrabutylammonium salts to 8×10^{-4} M.

Tables 3, 4, and 5 summarise the data for the MLCT band obtained in pure DMSO and with addition of tetra-butyl ammonium chloride and dihydrogen phosphate salts to the complexes. All the complexes show a change in absorbance on the addition of dihydrogen phosphate, whereas only the MLCT band of complex **3** appears to be sensitive to the binding of chloride.

Table 3 Electronic absorption data in DMSO on the addition of Cl^- and H_2PO_4^- to **1**^a

$\lambda_{\text{max}}/\text{nm}$ ($10^{-4}\epsilon/\text{M}^{-1}\text{cm}^{-1}$)			
1	Zero addition	Cl^-	H_2PO_4^-
$\pi-\pi^*$ (1)	292 (10.50)	292 (10.9)	292 (11.18)
MC	358 (1.40)	358 (1.27)	358 (1.25)
MLCT	470 (2.45)	470 (2.45)	470 (2.85)

^a Absorption bands below 240 nm cannot be resolved from solvent. Solutions were 10^{-5} M.

Cl^- and H_2PO_4^- were added as tetrabutylammonium salts to 8×10^{-4} M.

Table 4 Electronic absorption data in DMSO on the addition of Cl^- and H_2PO_4^- to **2**^a

$\lambda_{\text{max}}/\text{nm}$ ($10^{-4}\epsilon/\text{M}^{-1}\text{cm}^{-1}$)			
2	Zero addition	Cl^-	H_2PO_4^-
$\pi-\pi^*$ (1)	292 (12.01)	292 (12.65)	292 (12.25)
MC	354 (1.74)	354 (1.76)	354 (1.57)
MLCT	466 (2.44)	466 (2.45)	466 (2.54)

^a Absorption bands below 240 nm cannot be resolved from solvent. Solutions were 10^{-5} M.

Cl^- and H_2PO_4^- were added as tetrabutylammonium salts to 8×10^{-4} M.

Table 5 Electronic absorption data in DMSO on the addition of Cl^- and H_2PO_4^- to **3**^a

$\lambda_{\text{max}}/\text{nm}$ ($10^{-4}\epsilon/\text{M}^{-1}\text{cm}^{-1}$)			
3	Zero addition	Cl^-	H_2PO_4^-
$\pi\text{-}\pi^*$ (1)	292 (31.46)	292 (29.53)	292 (26.85)
MC	*	*	*
MLCT	460 (8.07)	462 (7.12)	464 (7.32)

^a Absorption bands below 240 nm cannot be resolved from solvent. Solutions were 4×10^{-6} M. Cl^- and H_2PO_4^- were added as tetrabutylammonium salts to 3.2×10^{-4} M.

* not resolved

4.1.3 Fluorescence emission in DMSO

With the noted electronic absorption spectrochemical recognition in mind, fluorescence emission spectroscopic measurements were carried out to probe anion binding. This highly sensitive technique earlier proved to be useful in the investigation of metal cation binding by crown ether- and acyclic anion receptor containing ruthenium(II) polypyridyl complexes.^{68, 85} In real sample analyses, interference from other fluorescent species are likely to be negligible due to the relatively high wavelength MLCT emission band in ruthenium(II) polypyridyl complexes. As for the prototype, a broad low-energy emission band was observed for complexes **1**, **2**, and **3**.

However the λ_{max} of the complexes are red shifted when compared against the prototype. The data obtained in DMSO on the addition of Cl^- and H_2PO_4^- is

However the λ_{max} of the complexes are red shifted when compared against the prototype. The data obtained in DMSO on the addition of Cl^- and H_2PO_4^- is summarised in **Table 6**. Blue shifts in all the receptor λ_{max} values were observed on the addition of H_2PO_4^- , as expected no change in the fluorescence spectrum of $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ was observed. The largest effects were observed with complex **2** which showed a λ_{max} shift of 16nm. The quantum yields (**Table 6**) have been determined for **1** and **2**, where **2** showed a substantial increase in intensity on the addition of H_2PO_4^- . For example **Fig. 1** shows the dramatic effect which sequential additions of H_2PO_4^- has on the fluorescence emission of **2**. The increase in emission is accounted for by considering the structure of **2**. Prior to binding, the anion receptor sites in close proximity to the substituted bipyridyl are free to move via rotation etc., this lack of structural rigidity increases the chance of non-radiative decay. It has been found from the X-ray crystal structure of **2**, determined that the H_2PO_4^- anions coordinates via hydrogen bonds to the amide (CO-NH) and to the calix[4]arene hydroxyl groups.¹⁰⁷ This has the effect of increasing the structural rigidity in the supramolecular anion complex.

Fig. 1 - Fluorescence emission spectra of **2** (1×10^{-6} M) on the addition of H_2PO_4^- in DMSO

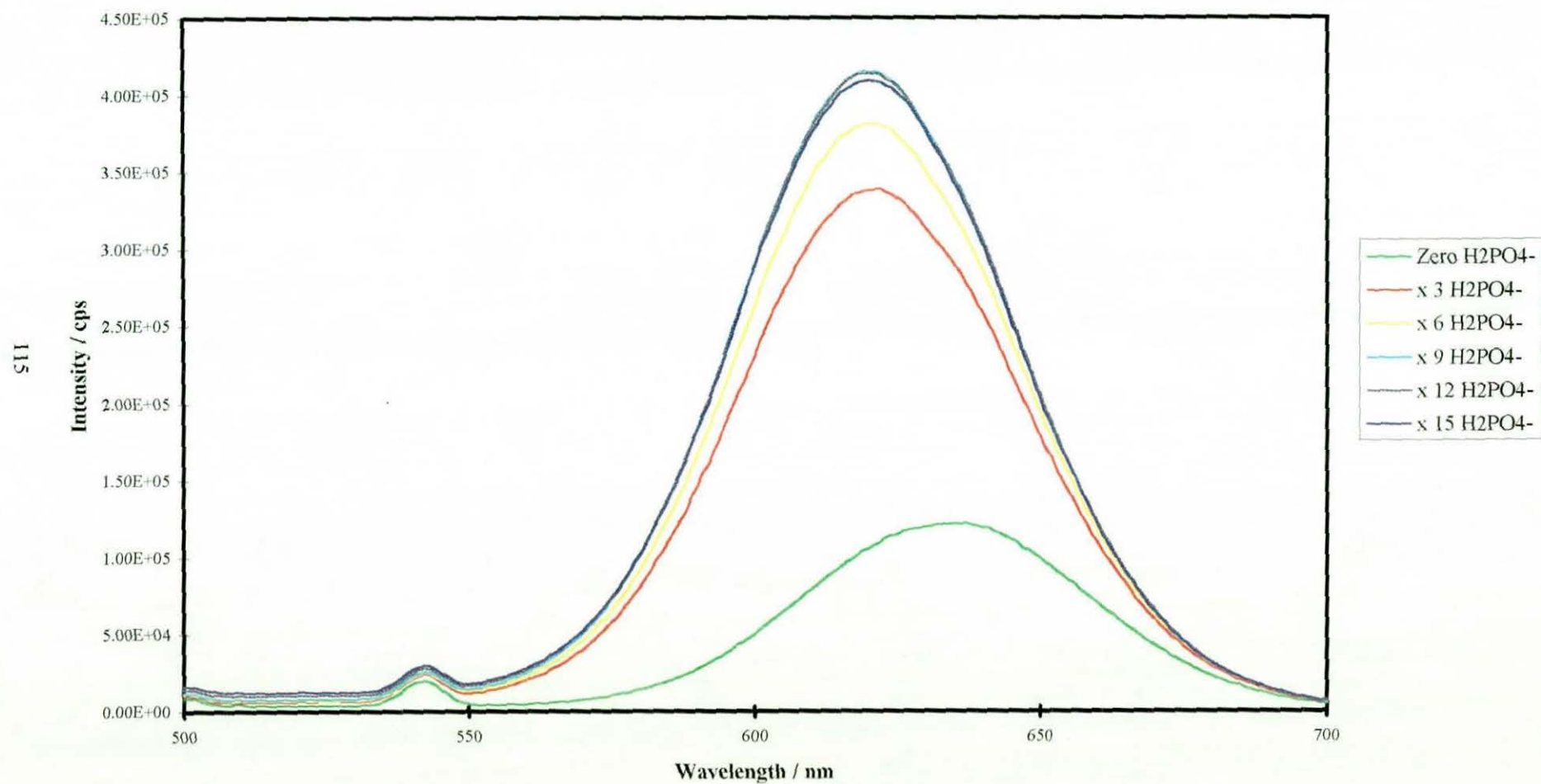


Table 6 Fluorescence emission data in DMSO on the addition of Cl^- and H_2PO_4^- to solutions of $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$, **1**, **2** and **3**^a

$\lambda_{\text{max}}/\text{nm}$ (quantum yield)			
	Pure DMSO	Cl^-	H_2PO_4^-
$[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$	607 (0.0620)	607 (0.0620)	607 ^b
1	638 (0.0184)	637 (0.0176)	635 (0.0240)
2	640 (0.0126)	638 (0.0099)	622 (0.0360)
3	628 ^c	626 ^c	622 ^c

^a Solutions were 10^{-6} M. Cl^- and H_2PO_4^- were added as tetrabutylammonium salts to 4×10^{-5} M.

^b Precipitation problems prevented quantum yield being accurately determined.

^c Quantum yield was not determined. Fluorescence was quenched on the addition of Cl^- and enhanced on the addition of H_2PO_4^-

Aliquot addition of anions to **2** was performed to determine the stability constants (K_s) of the complex on the addition of Cl^- and H_2PO_4^- . The stability constants (**determined using Formula 1 in Chapter 3 and see appendix 1 for derivation**) are detailed in **Table 7**, which shows that H_2PO_4^- forms a substantially more stable complex with **2** than Cl^- .^{110, 111} The selectivity for H_2PO_4^- over Cl^- was determined to be 11.5 using the **Formula 1**. As previously stated above the X-ray crystal structure of **2** shows that the H_2PO_4^- is coordinated via hydrogen bonds to the amide (CO-NH) and to the calixarene hydroxyl groups. This offers a possible explanation to the high selectivity because it is

reasonable to assume that the Cl^- anion due to its geometry will be coordinated by **2** hydrogen bonds.

Table 7. Stability constant for **2** in DMSO on the aliquot addition of Cl^- and H_2PO_4^-

Anion	$\log K_s$
Cl^-	4.10
H_2PO_4^-	5.15

Formula 1

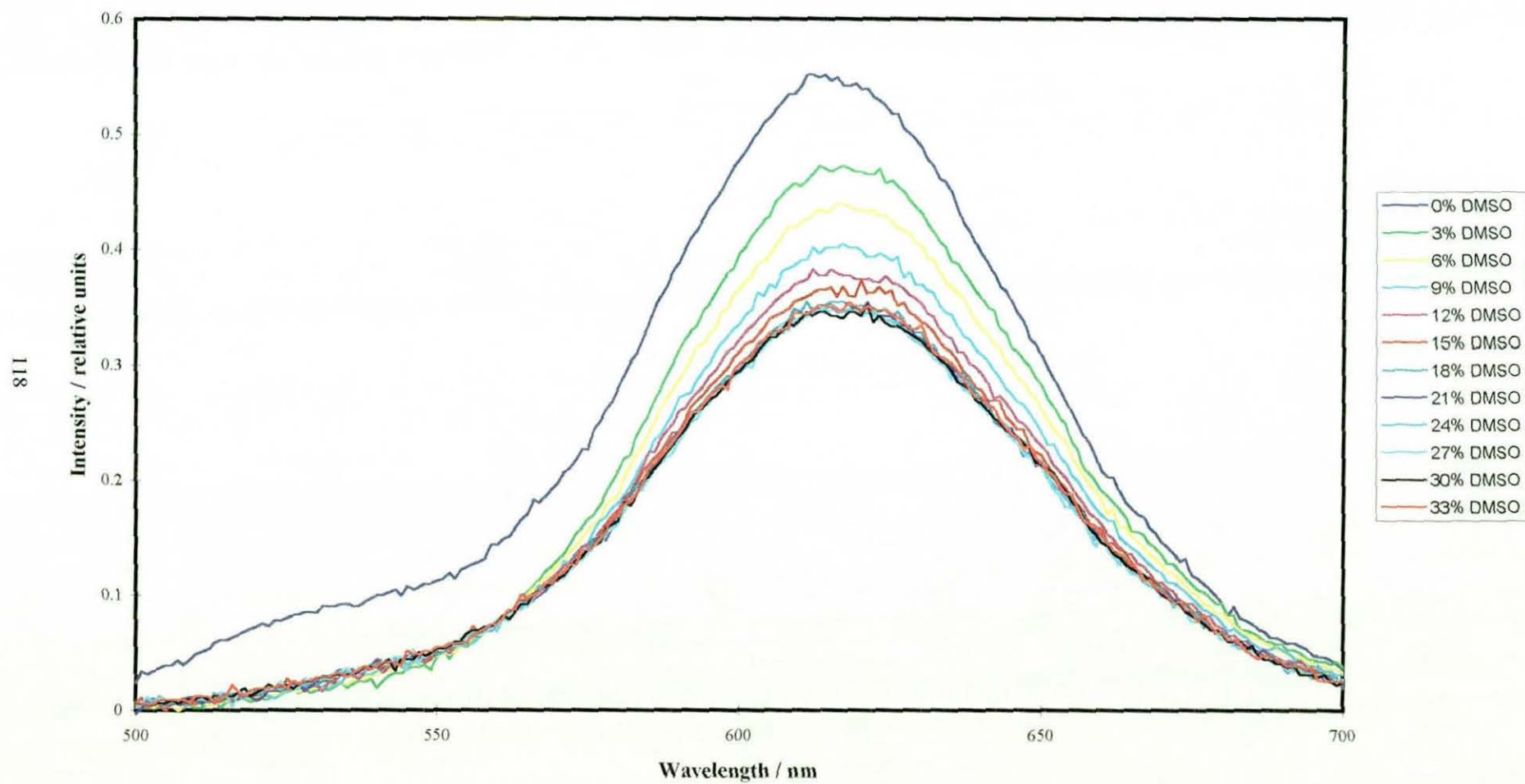
$$\text{Selectivity} = \text{Stability Constant } (\log K_s \text{ H}_2\text{PO}_4^-) / \text{Stability Constant } (\log K_s \text{ Cl}^-)$$

4.2 Solvent complexation studies

4.2.1 Emission spectroscopy in acetonitrile on the addition of DMSO to **3**

The work in Chapter 3 - section 3.1.2 indicated that a polar solvent such as DMSO would solvate the cavity created for the anion. This may lead to the solvent competing with anions for the binding site. A study of the effect of the solvent on the receptor was conducted using fluorescence emission spectroscopy by making aliquot additions of DMSO (0% to 25%) to **3** in acetonitrile. The fluorescence spectra **Fig. 2** detail the effect of the aliquot additions of DMSO. On increased additions of DMSO the

Fig. 2 - Fluorescence emission spectra of **3** (1×10^{-6} M) on the addition of DMSO in acetonitrile



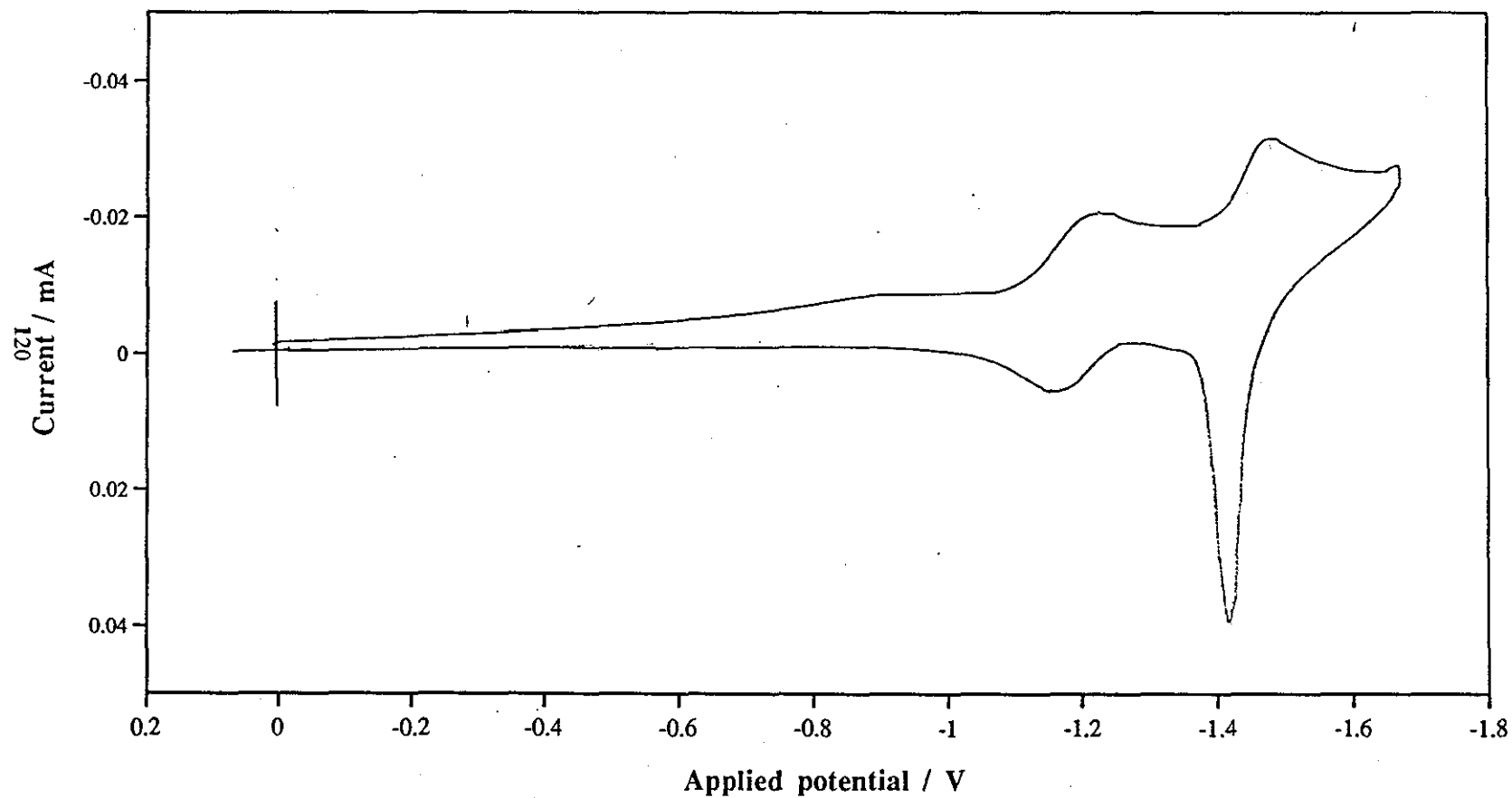
fluorescence is rapidly quenched and there is a small red shift in the λ_{max} of 3nm. This implies that DMSO solvates the cavity which leads to the quenching in the fluorescence.

4.3 Electrochemical complexation study

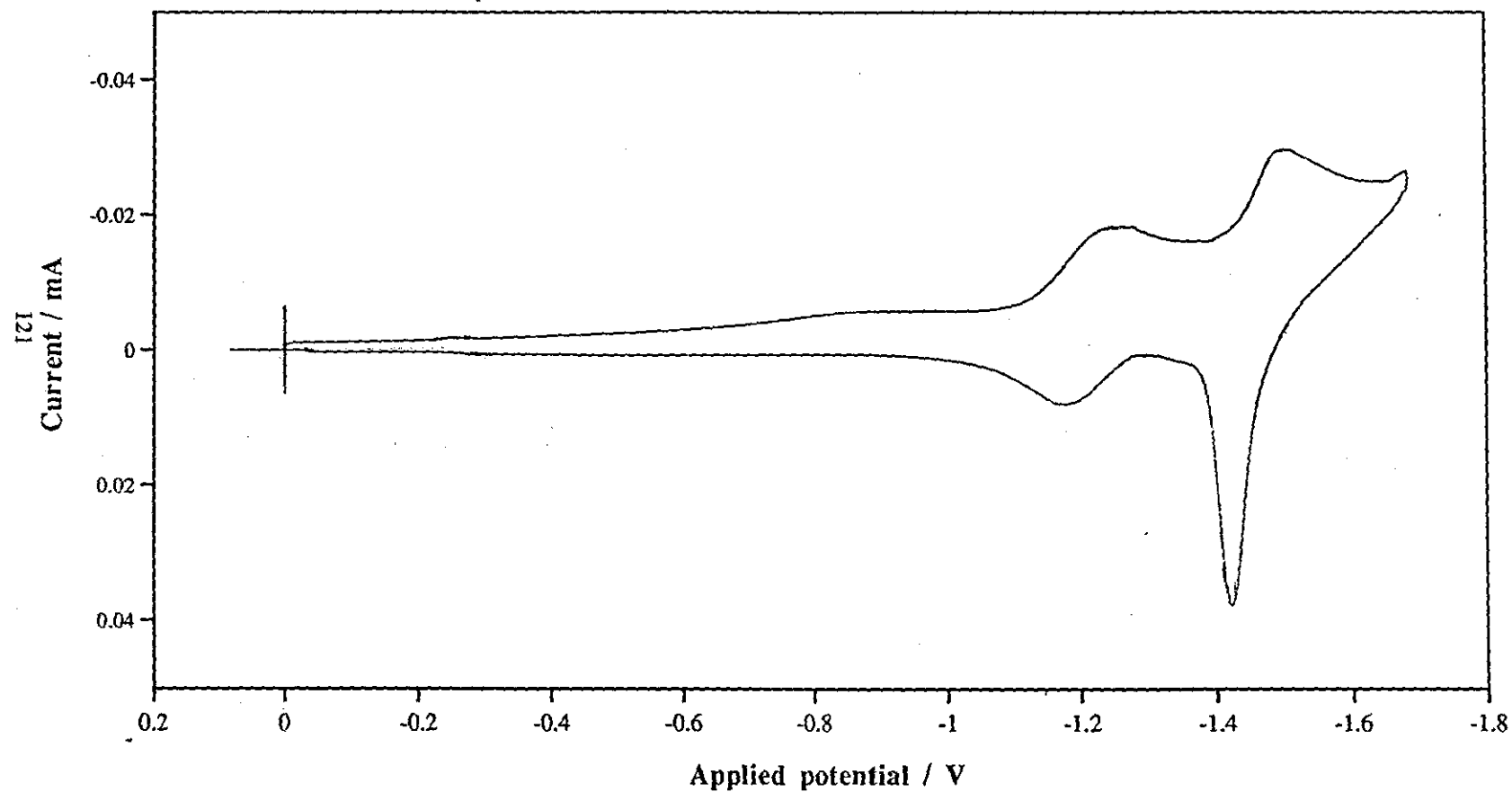
4.3.1 Cyclic voltammetry

As previously mentioned the tris-2,2'-bipyridyl ruthenium(II) complexes undergoes a metal-centred oxidation process and a series of three reduction waves corresponding to the successive one-electron reduction of the three ligands. Beer et al. have previously reported electrochemical measurements on **2**.¹¹⁷ They showed that substantial anion-induced cathodic perturbations of the respective ligand-centred amide substituted bipyridyl reduction redox couple were detected in electrochemical anion recognition experiments with **2**. These electrochemical experiments showed that the H_2PO_4^- forms a more stable complex than Cl^- , which is in agreement with emission and UV-vis spectroscopy. Here we report the electrochemical anion recognition experiments performed on $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$ and **3** (Tables 8 and 9). It was not possible to study electrochemical changes on the addition of H_2PO_4^- due to precipitation of both complexes. Electrochemistry of the prototype molecule showed no changes on the addition of Cl^- as given in Table 8. The electrochemistry of **3** is very interesting, where the CVs **1**, **2**, **3** and **4** shows that on scanning to the second ligand redox wave the neutral form is adsorbed. It is reasonable to assume that the effect occurs because of the low solubility of the neutral species in acetonitrile due to the incorporation of the

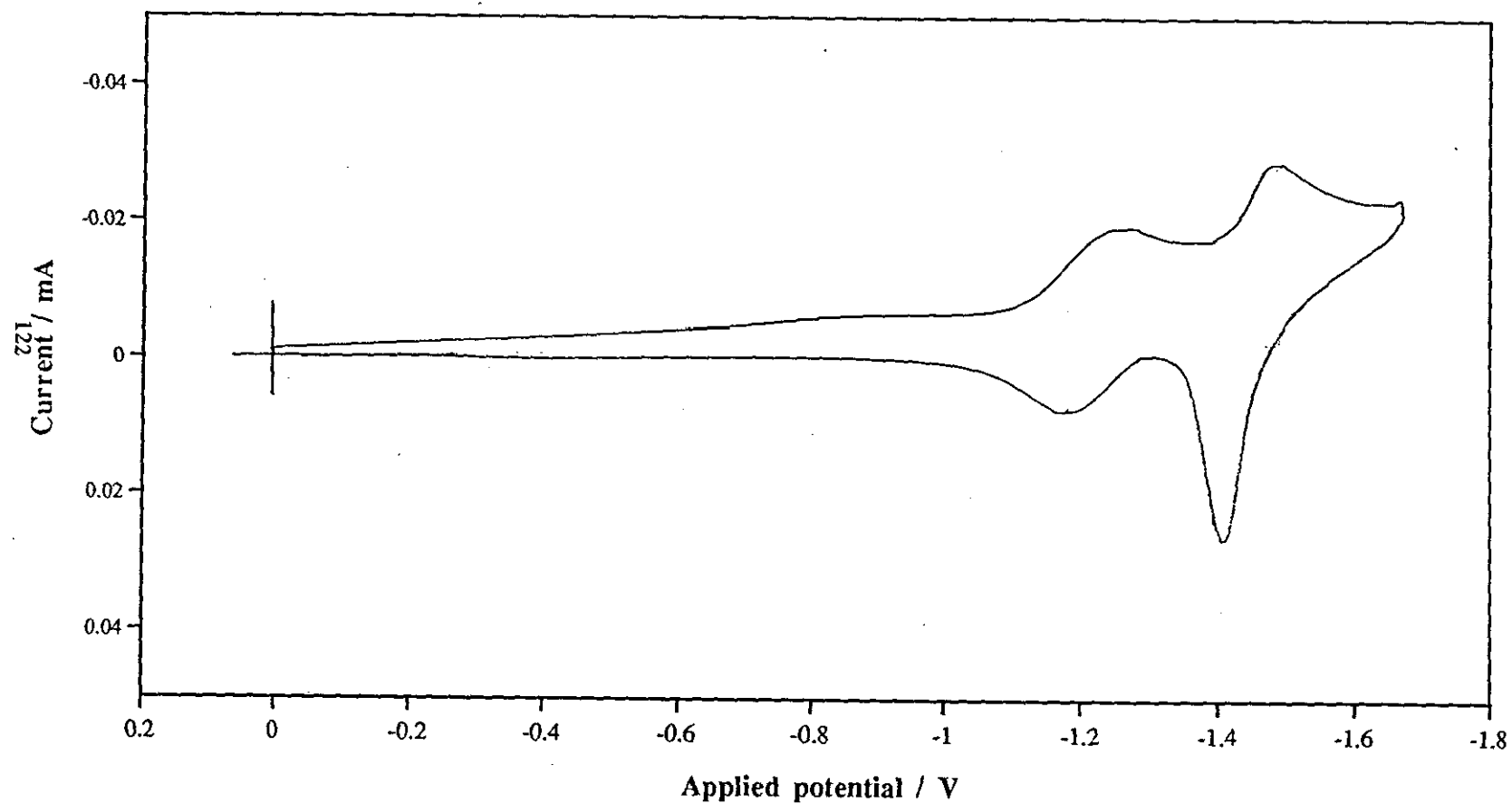
CV 1 - CV at 100 mVs^{-1} in an acetonitrile solution containing 0.2 mM of 3 in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode.



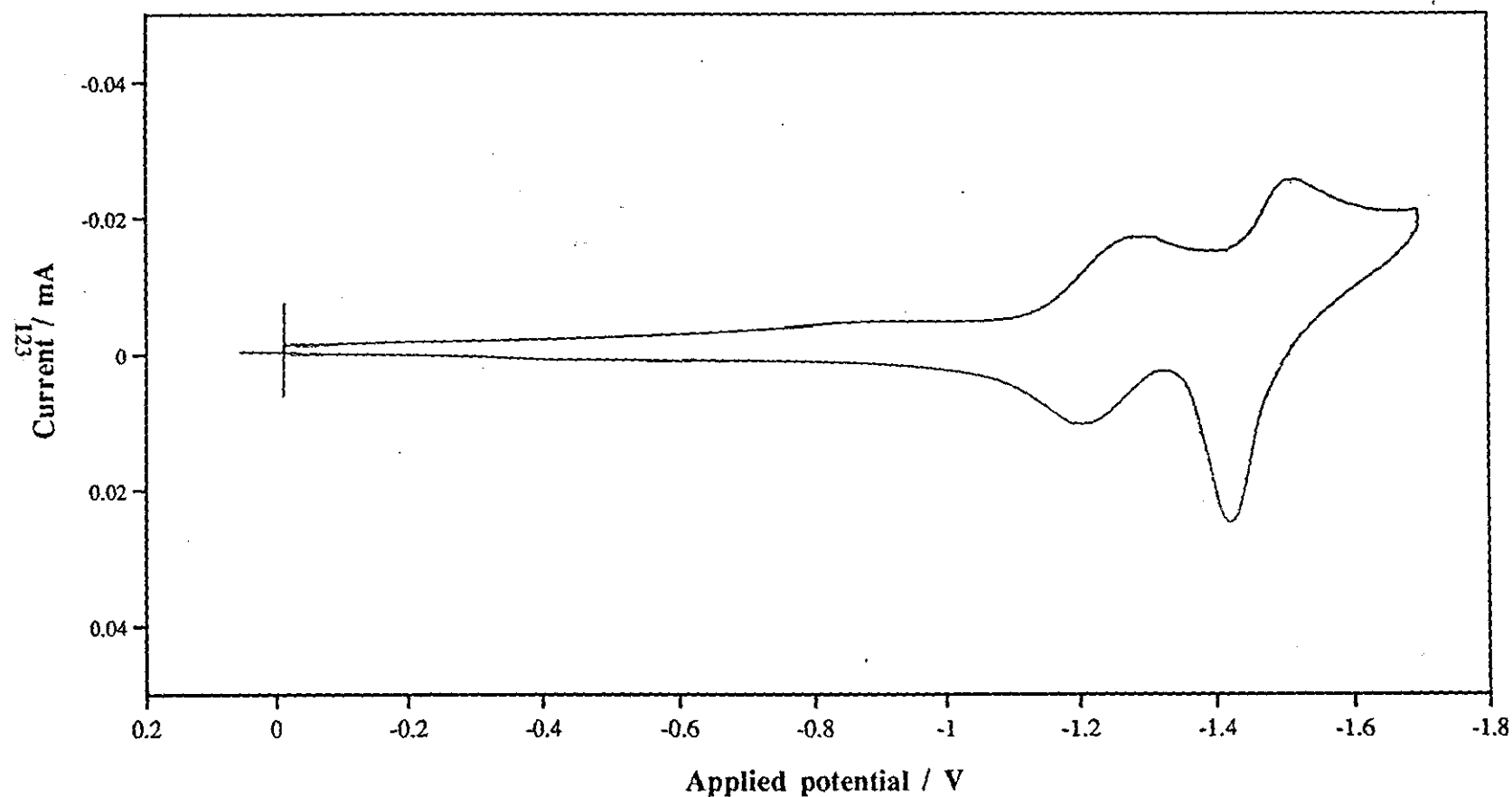
CV 2 - CV at 100 mVs^{-1} in an acetonitrile solution containing 0.2 mM of 3 on the addition of 0.4 mM tetrabutylammonium chloride in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode.



CV 3 - CV at 100 mVs^{-1} in an acetonitrile solution containing 0.2 mM of 3 on the addition of 2 mM tetrabutylammonium chloride in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode.



CV 4 - CV at 100 mVs^{-1} in an acetonitrile solution containing 0.2 mM of 3 on the addition of 4 mM tetrabutylammonium chloride in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode.



calix[4]arene moiety. A number of diagnostic tests were performed on the proposed anodic stripping wave. Scan rate dependencies and anodic peak potentials and currents were compared against scan rates. The scan rates showed direct linearity when compared against the anodic peak currents and was found to be independent of the anodic peak current. This further supports that this wave is due to anodic stripping of the neutral species of the electrode.

Table 8 Electrochemical data for $[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]^a$

$[\text{Ru}(\text{bipy})_3(\text{PF}_6)_2]$	$E^f(+2/+1)$	$E^f(+1/0)$	$E^f(0/-1)$	$E^f(-1/-2)$
Zero Chloride	-1.33	-1.53	-1.46	-1.27

^a Obtained in acetonitrile solution containing 0.1 M Bu_4NBF_4 as supporting electrolyte.

Solutions were 2×10^{-4} M in receptor, and potentials were determined with reference to a saturated sodium chloride calomel electrode at 25 ± 2 °C at 100 mV s^{-1} scan rate.

Table 9 Electrochemical data for **3**^a

3	$E^f(+2/+1)$	$E^f(+1/0)$	$E^f(0/-1)$	$E^f(-1/-2)$
Zero Chloride	-1.21	-1.46	-1.39 (40 μA)	-1.15
*2 Chloride	-1.23	-1.47	-1.39 (39 μA)	-1.15
*10 Chloride	-1.24	-1.47	-1.38 (28 μA)	-1.16
*20 Chloride	-1.24	-1.47	-1.38 (23 μA)	-1.17

^a Obtained in acetonitrile solution containing 0.1 M Bu_4NBF_4 as supporting electrolyte.

Solutions were 2×10^{-4} M in receptor, and potentials were determined with reference to a saturated sodium chloride calomel electrode at 25 ± 2 °C at 100 mV s^{-1} scan rate.

As with the previously studied complexes (Chapter 3), the least cathodic bipyridyl ligand centred reduction couple can be assigned to the amide-substituted bipyridyl group due to the electron-withdrawing nature of the carbonyl amide moiety. This redox wave undergoes a cathodic shift of 30mV on the addition of Cl^- , which is due to an increased electron density in the environment of the receptor site. The stripping wave also broadened and a decrease in the peak current on the increasing addition of Cl^- is observed (Table 9). Changes in the nature of this stripping wave on increased addition of Cl^- may be due to geometrical effects or the negative charge on anion which leads to alterations in the electron transfer kinetics.

4.4 Conclusion

The Lewis acidic redox-active and photo-active ruthenium (II) bipyridyl moiety in combination with amide CO-NH groups has been successfully incorporated into macrocyclic and calix[4]arene structural frameworks to produce novel receptors capable of the electrochemical and spectral recognition of anions. As noted with the complexes in Chapter 3, electronic adsorption and emission spectroscopy measurements on the complexes **1-3** have shown that these receptors are sensitive to the binding of anions such as Cl^- and H_2PO_4^- . In contrast to the acyclic receptors (Chapter 3) the spectroscopic studies of macrocyclic and calix[4]arene receptor molecules with increased rigidity were performed in DMSO rather than acetonitrile. Measurements in this more polar solvent were possible due to increased rigidity of the anion receptor site which prevented the molecule from being extensively distorted due

to solvation. Emission studies on the complexes in DMSO showed significant perturbations in λ_{max} and quantum yields on the addition of Cl^- and H_2PO_4^- . The most dramatic change was in the emission properties of **2** which showed a near tripling in the quantum yield and a shift in λ_{max} of 18 nm to lower energy.

Electrochemical anion recognition studies were performed on complex **3** which showed a cathodic shift of 30 mV in the amide-substituted bipyridyl group on the addition of Cl^- . A stripping peak was observed when cyclic voltammetric experiments were performed on complex **3**, which indicated that the neutral form of the receptor undergoes surface adsorption onto the electrode. The shape of this stripping peak changed on the addition of Cl^- , which may be due to an alteration in the electron transfer kinetics.

Chapter 5 - Diquat Based Receptors

5.0 Introduction

In chapters 3 and 4 ruthenium (II) trisbipyridyl derivatives have been used in the complexation of anions such as in Fig. 1. As well as the ruthenium metal centre being a Lewis acid, the combination with the bipyridyl groups rigidifies the receptor sites. This prevents rotation of the bond joining the pyridine groups, which would be detrimental to the formation of strong complexes. A number of these $[\text{Ru}(\text{bipy})_3]^{2+}$ based receptors have proved to be complexing agents for cations and anions.^{85, 96, 97}

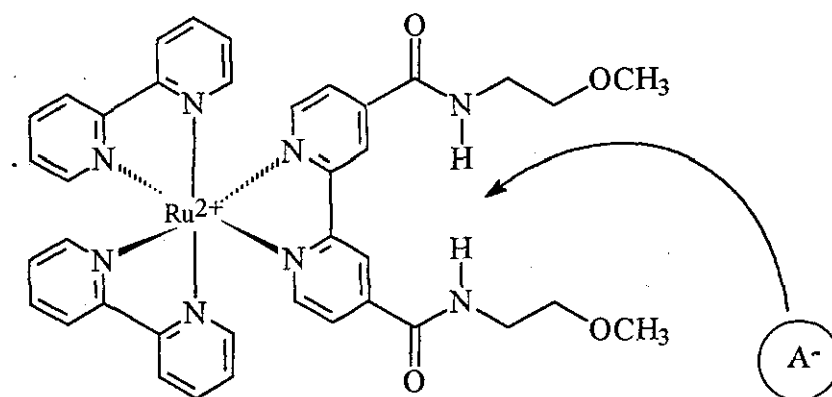


Fig. 1 Example of a ruthenium(II) trisbipyridyl receptor for anions

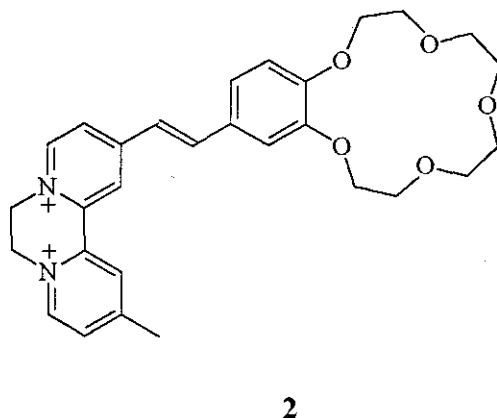
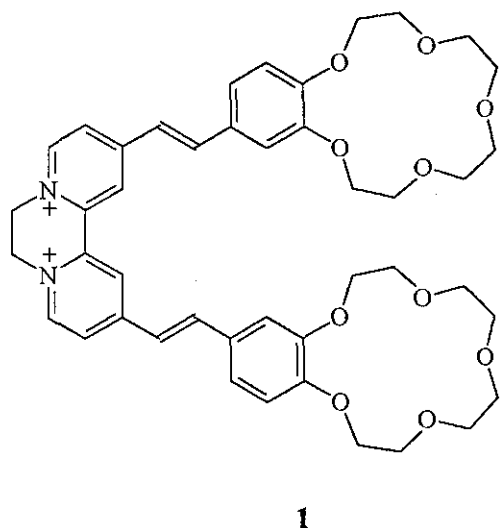
Quaternisation of the bipyridyl ligands to prepare the diquat¹¹⁸ (systematic name for diquat is 6,7-dihydrodipyrido[1,2-a: 2',1'-c]pyrazidinium) derivatives offers an alternative and possibly advantageous approach to $[\text{Ru}(\text{bipy})_3]^{2+}$ derivatives as:-

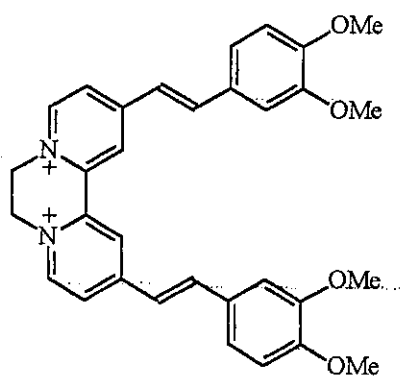
1. synthesis of these $[\text{Ru}(\text{bipy})_3]^{2+}$ derivatives is complicated in comparison with that of diquat

2. starting material costs are also greater due to the cost of the ruthenium metal

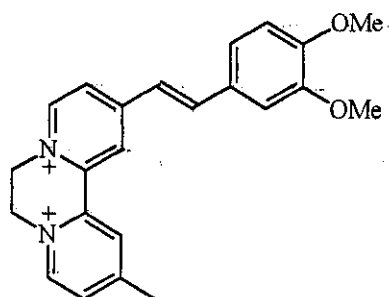
This gave the impetus to conduct research into the cation and anion complexing ability of diquat based derivatives. Details of the synthesis of diquat **5** and the substituted diquat based derivatives **1-4** are given in the experimental section.

The bipyridylium salts have a number of interesting properties such that diquat dibromide readily gives rise to stable cation radicals by a one-electron reduction process.¹¹⁹ Diquat has interesting herbicidal properties, which appear to be due to the ease of (reversible) one-electron reduction to form stable but air-sensitive cation radicals. Research has indicated that the mechanism of the reaction as a herbicide is due to the reduction of the diquat dication by light. The formation of this radical cation is toxic to the plant when it is absorbed. From these previous physical properties it is envisaged that the complexation of cations and anions by **1-5** can be probed in a similar fashion to the $[\text{Ru}(\text{bipy})_3]^{2+}$ derivative **6** using electrochemistry, UV-vis absorbance and ^1H NMR spectroscopy.

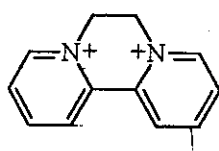




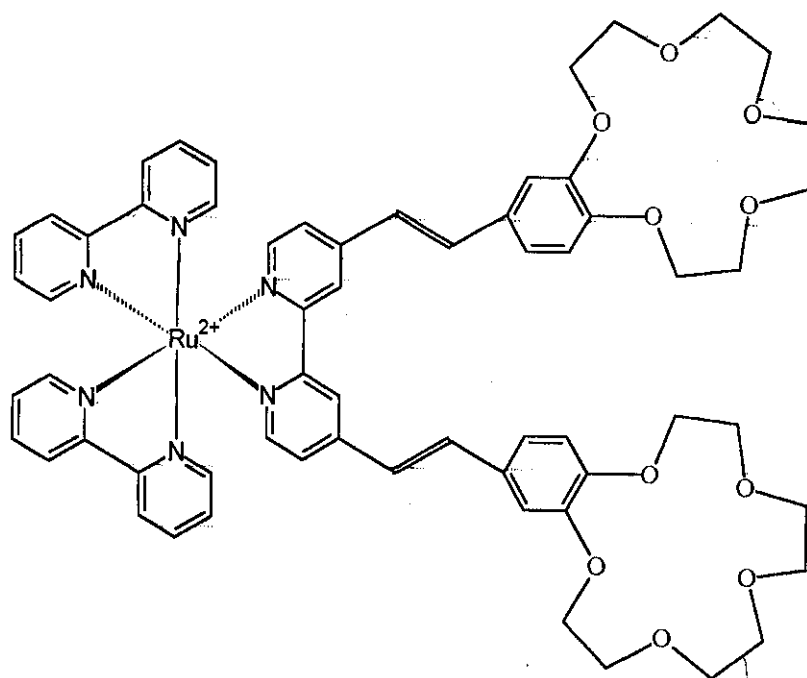
3



4



5



6

5.1 Metal Cation Complexation studies

5.1.1 UV-vis spectroscopy

Table 1, absorbance data of **1**, **2**, **5** and benzo-15-crown-5, demonstrate the electronic communication that takes place on the linking of benzo-15-crown-5 and **5** which give the resulting absorbances for **1** and **2**. The extra **1** and **2** absorbances are due to the incorporation of the vinylic groups within the diquat derivative, which increases the conjugation within the molecules. This results in absorption within the visible region for the diquat derivatives **1-4** due to the increased conjugation, where the diquat derivatives **1-4** are red in colour whereas the diquat and benzo-15-crown-5 are white. These colours are attributed to charge transfer between the electron-rich catechol units of the molecules **1-4** and the electron deficient bipyridinium ring system of the diquat dication.

Table 1 Electronic absorption data for **5**, benzo-15-crown-5, **1** and **2**^a in acetonitrile

$\lambda_{\text{max}}/\text{nm}$ ($10^{-4}\epsilon/\text{M}^{-1}\text{cm}^{-1}$)			
5 (Diquat)	Benzo-15-crown-5	1	2
250 (0.57)	276 (0.78)	264 (2.47)	254 (3.76)
308 (2.57)		322 (3.24)	316 (2.09)
406 (0.05)		356 (3.43)	350 (1.15)
		452 (4.52)	458 (2.36)

^a Absorption bands below 240 nm cannot be resolved from solvent. Solutions concentrations for **5** and benzo-15-crown-5 were 6×10^{-5} M and 2×10^{-5} M for **1** and **2**

On separate addition of excess Mg^{2+} , Na^{+} and Li^{+} cations to **1** and **2** solutions the most significant spectral change was in the visible region. The absorbance bands at 452 and 458 nm (**Tables 2** and **3**) for **1** and **2**, respectively, undergo large hypsochromic shifts. It is reasonable to postulate that each positively charged metal cation on complexation with the electron rich benzo-15-crown-5 moiety reduces its ability to undergo charge transfer. As expected the Mg^{2+} cation causes the largest λ_{max} shifts in **1** (42nm) and **2** (36nm) due to it having the highest polarising power. With these large λ_{max} shifts the solutions of **1** and **2** undergo colour changes on the addition of all the cations. **1** and **2** show colour changes on the addition of Mg^{2+} , Na^{+} and Li^{+} to dark yellow, yellow/orange and yellow/orange respectively from the original orange colouration. Spectra of **1** on the aliquot addition of Mg^{2+} , Na^{+} and Li^{+} cations is shown in **Figs. 2, 3**,

Fig. 2 - Electronic absorption spectra of **1** (2×10^{-5} M) on the addition of Mg^{2+} (excess wrt. molecule) in acetonitrile

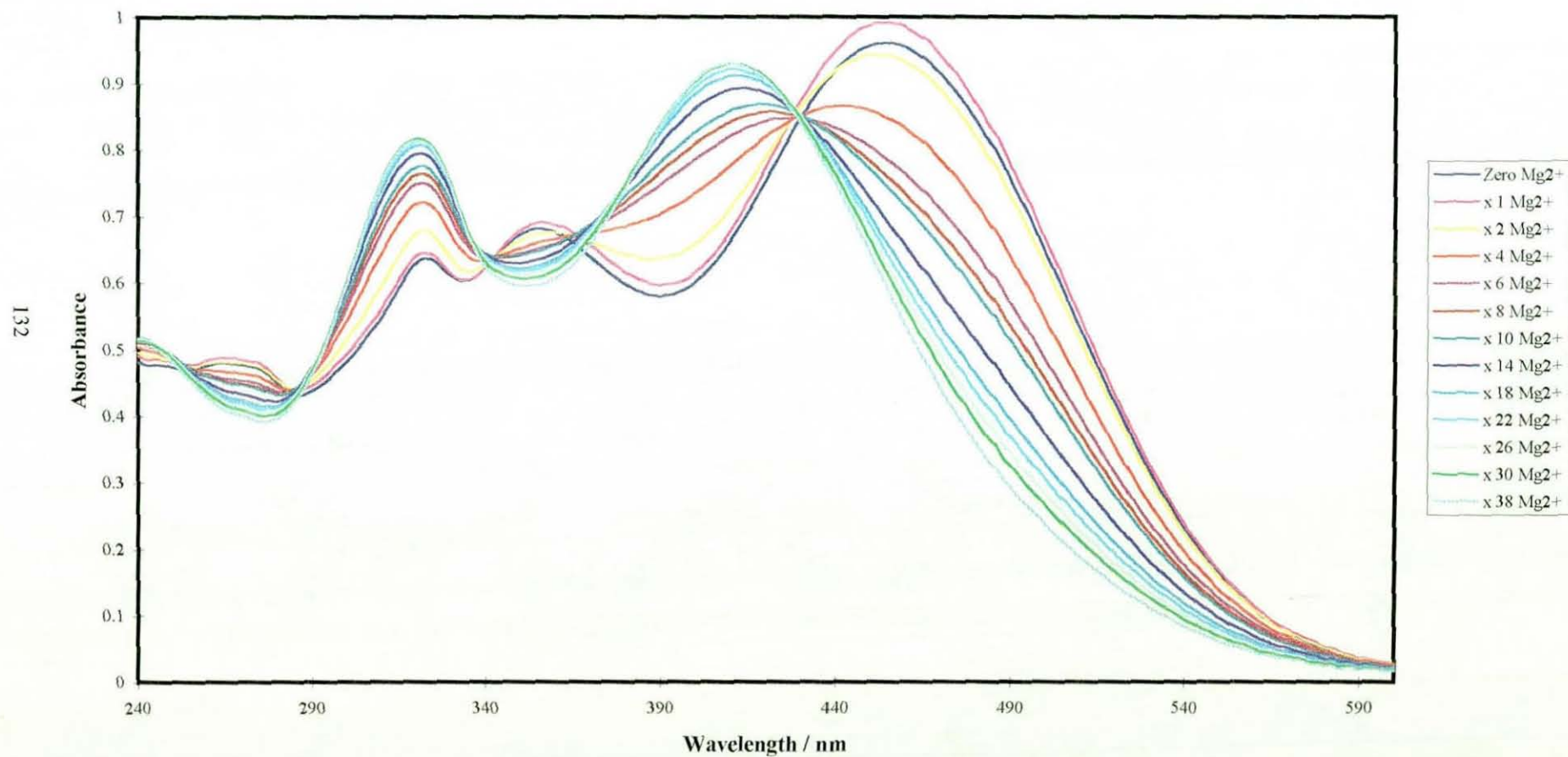
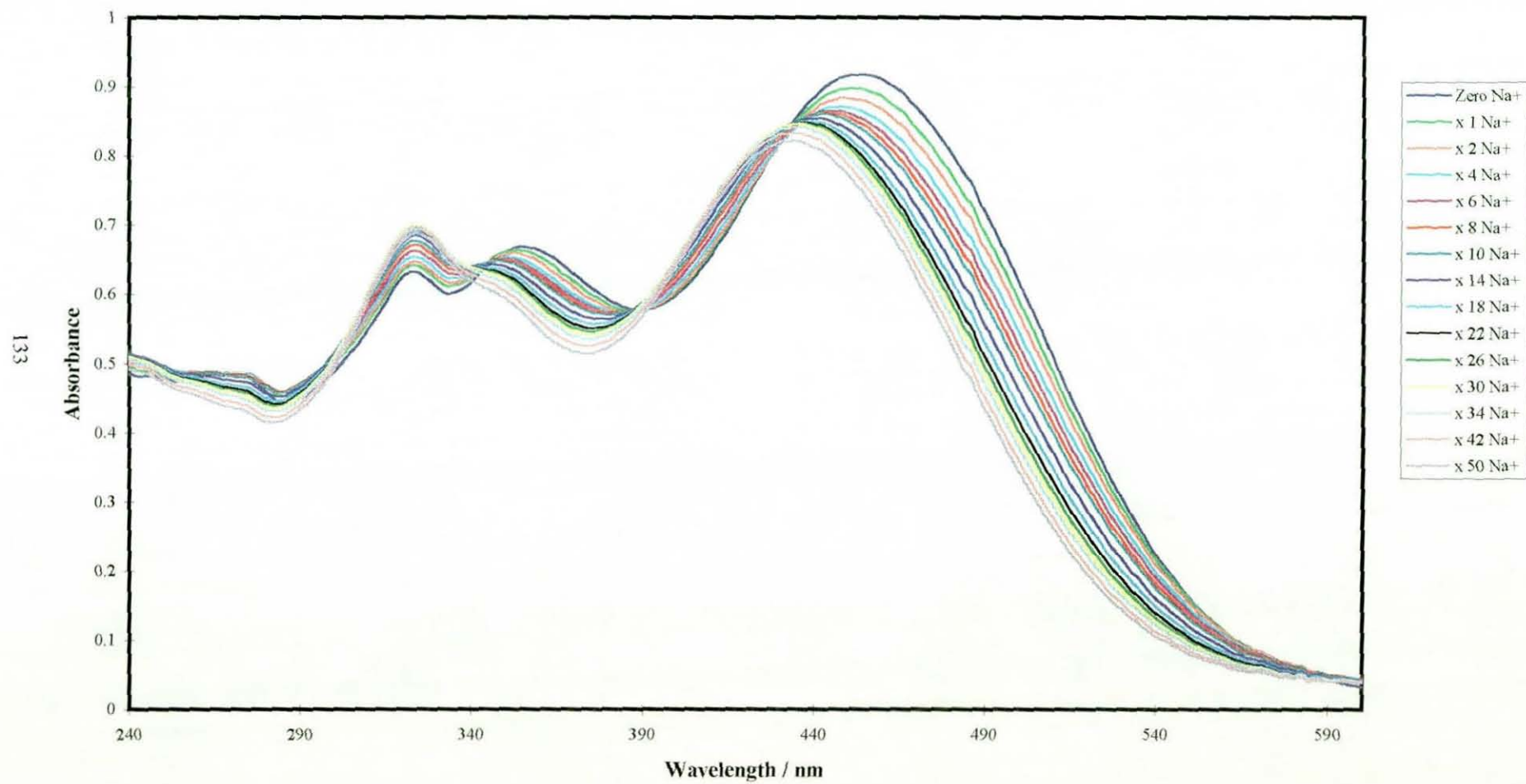


Fig. 3 - Electronic absorption spectra of **1** (2×10^{-5} M) on the addition of Na^+ (excess wrt molecule) in acetonitrile



and **4** respectively. Similar effects are seen on the aliquot addition of these cations to **2** (Figs. 5, 6 and 7).

As expected, the bismethoxy diquat derivatives **3** and **4** without the benzo-15-crown-5 groups showed negligible effects in their absorption spectra on the addition of excess Mg^{2+} , Na^+ and Li^+ cations. As noted above, **1** and **2** both showed changes in their absorbance spectra on the addition of Mg^{2+} , Na^+ and Li^+ .

Table 2 Electronic absorption data for **1** in acetonitrile on the addition of Mg^{2+} , Na^+ and Li^+ ^a

$\lambda_{\text{max}}/\text{nm} (10^{-4}\epsilon/\text{M}^{-1} \text{ cm}^{-1})$			
Pure Solvent	Mg^{2+}	Na^+	Li^+
264 (2.47)	240 (2.54)	*	*
322 (3.24)	320 (4.17)	316 (3.60)	324 (3.88)
354 (3.43)	*	*	*
452 (4.52)	410 (4.37)	434 (4.17)	438 (4.67)

^a absorption bands below 240 nm cannot be resolved from solvent. Solution concentrations were 2×10^{-5} M. Mg^{2+} , Na^+ and Li^+ were added as perchlorate salts to 1.2×10^{-3} M

* Merges into neighbouring peak

Fig. 4 - Electronic absorption spectra of **1** (2×10^{-5} M) on the addition of Li^+ (excess wrt molecule) in acetonitrile

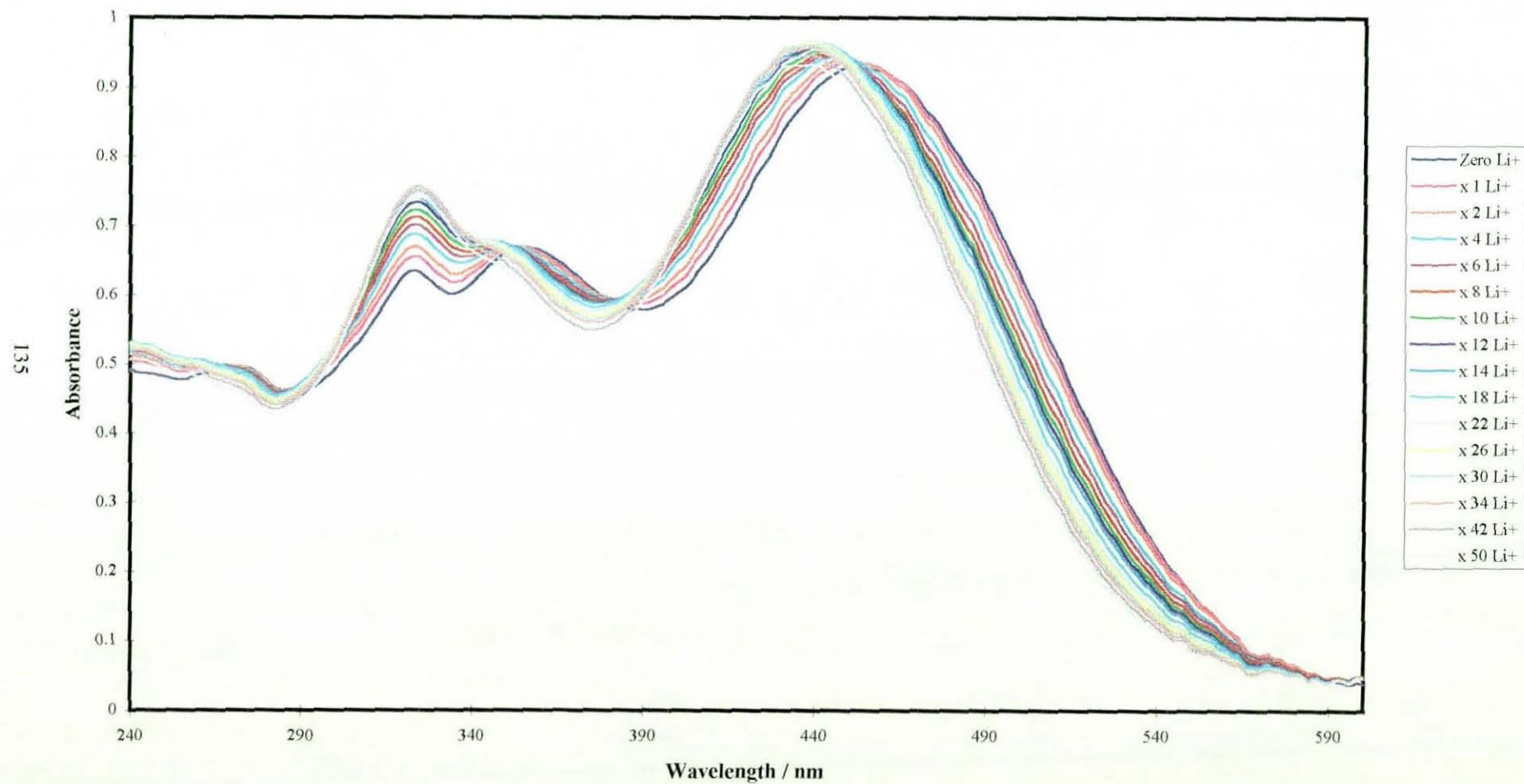


Fig. 5 - Electronic absorption spectra of **2** (2×10^{-5} M) on the addition of Mg^{2+} in acetonitrile

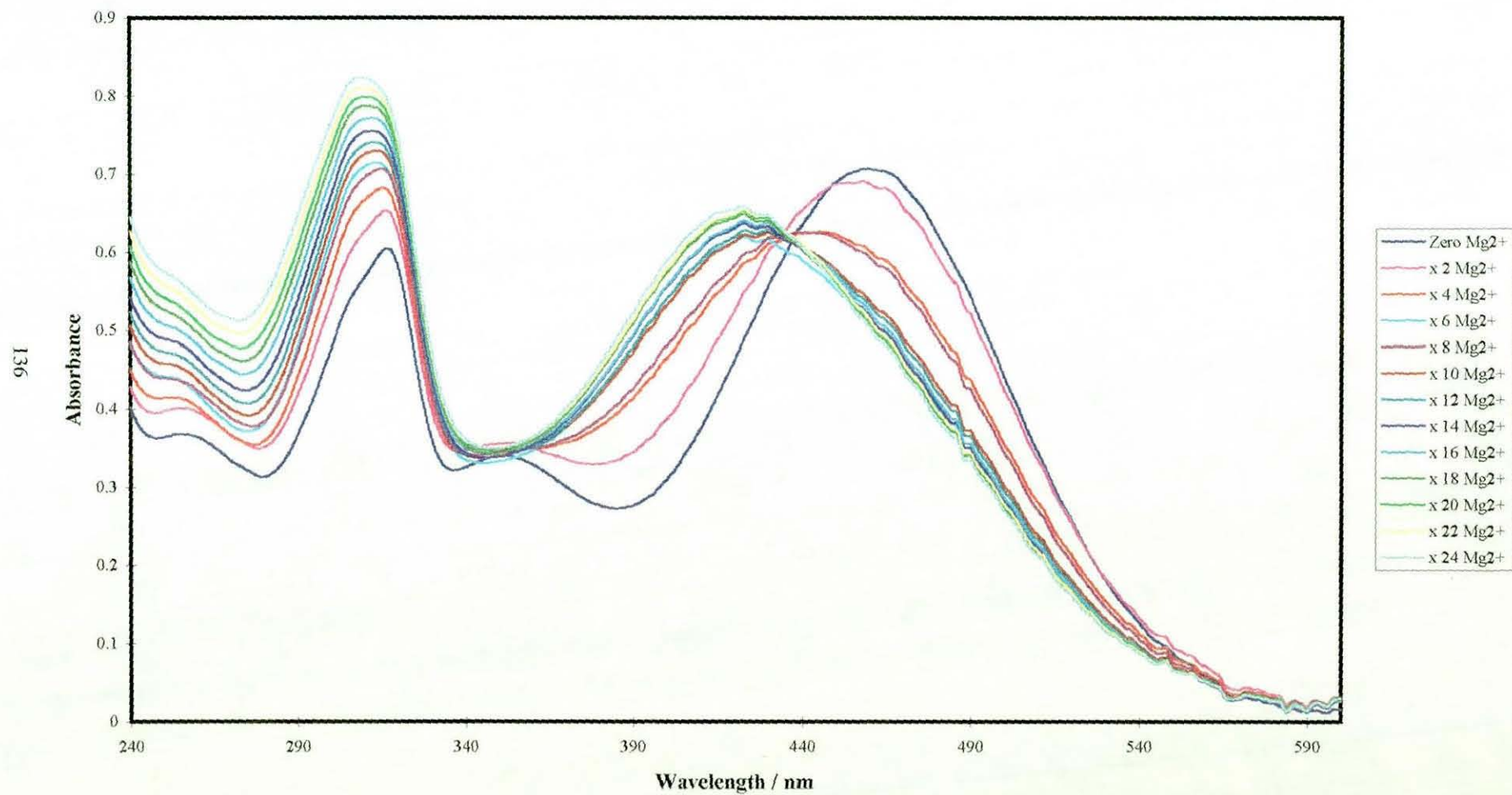


Fig. 6 - Electronic absorption spectra of **2** (2×10^{-5} M) on the addition of Na^+ in acetonitrile

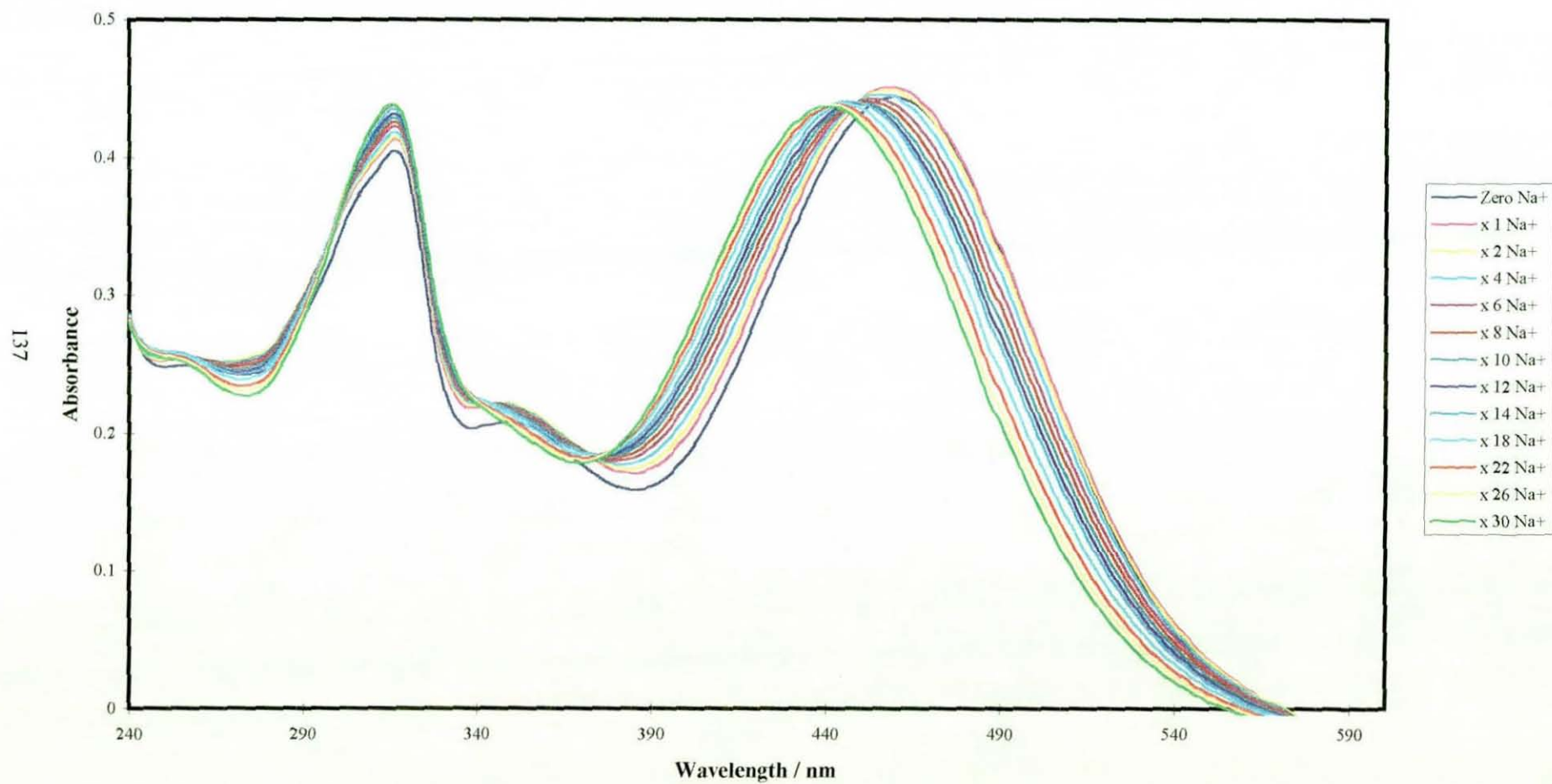


Fig. 7 - Electronic absorption spectra of **2** (2×10^{-5} M) on the addition of Li^+ in acetonitrile

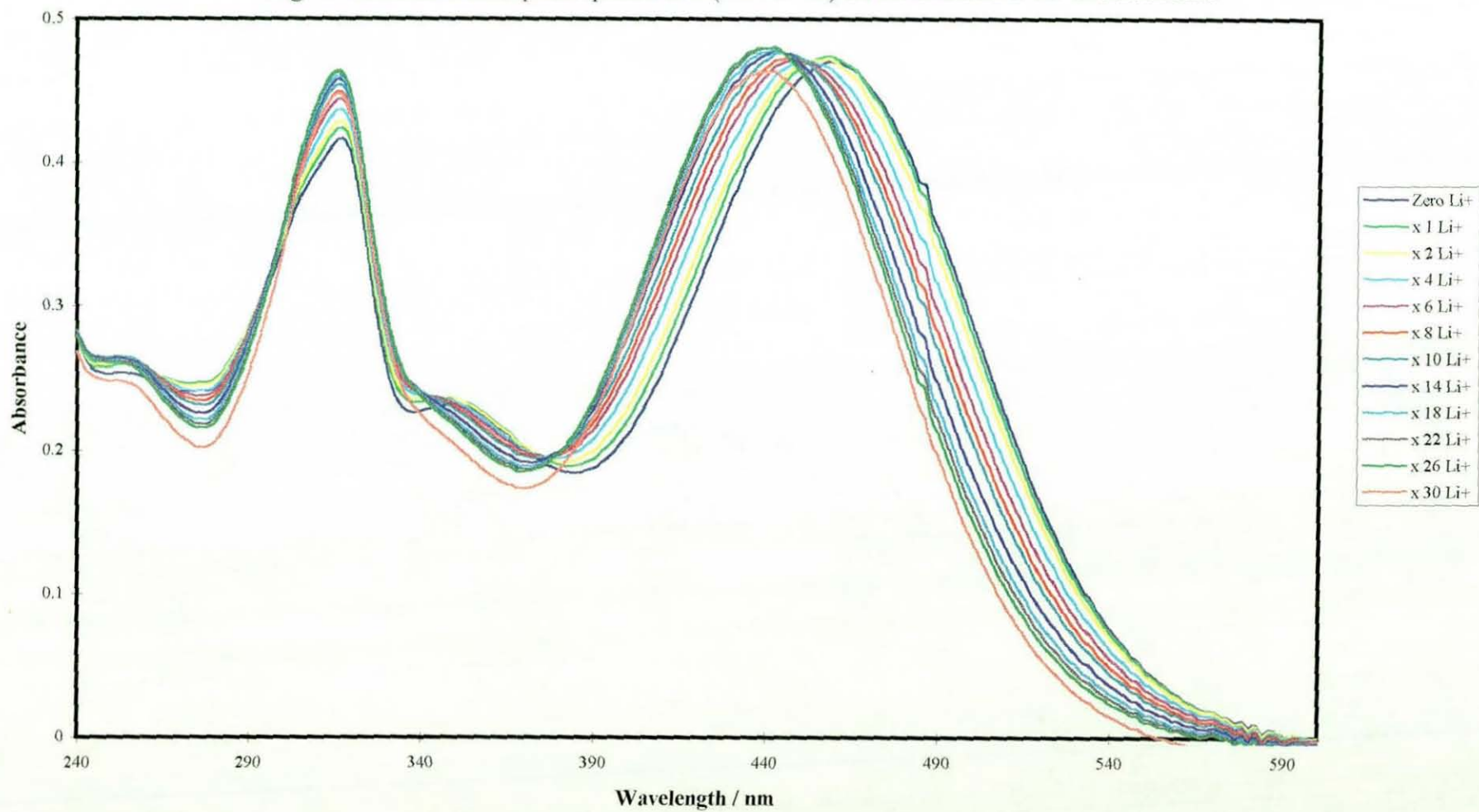


Table 3 Electronic absorption data for **2** in acetonitrile on the addition of Mg^{2+} , Na^+ and Li^+ ^a

$\lambda_{\text{max}}/\text{nm} (10^{-4}\epsilon/\text{M}^{-1} \text{ cm}^{-1})$			
Pure Solvent	Mg^{2+}	Na^+	Li^+
254 (1.27)	*	*	*
316 (2.09)	312 (2.52)	316 (2.27)	316 (2.33)
350 (1.15)	*	*	*
458 (2.36)	422 (2.13)	442 (2.33)	442 (2.42)

^a absorption bands below 240 nm cannot be resolved from solvent. Solution concentrations were 2×10^{-5} M. Mg^{2+} , Na^+ and Li^+ were added as perchlorate salts to 6×10^{-4} M

* Merges into neighbouring peak

Benzo-15-crown-5 was studied using UV-vis spectroscopy on the addition of 24 x excess of Mg^{2+} . Changes in the absorbances and λ_{max} were noted on the addition of Mg^{2+} (Table 4).

Table 4 Electronic absorption data for benzo-15-crown-5 in acetonitrile on the addition of Mg^{2+} ^a

$\lambda_{\text{max}}/\text{nm} (10^{-4}\epsilon/\text{M}^{-1} \text{ cm}^{-1})$	
Benzo-15-crown-5	Mg^{2+}
276 (0.78)	268 (0.74)

^a absorption bands below 240 nm cannot be resolved from solvent. Solution concentrations were 6×10^{-5} M. Mg^{2+} was added as perchlorate salts to 1.8×10^{-3} M

These changes in magnitude were smaller than those obtained for **1** and **2** (Tables 2 and 3). This indicates that the incorporation of the vinyl group into **1** and **2** results in the extension of the spectral range of the benzo-15-crown-5.

The stability constant K_s which controls the equilibrium between the free ligand L and the complex ML with the metal M was obtained from the variations of absorbance of **1** and **2** with aliquot addition of cations.

Stability constants were calculated using **Formula 1** (see appendix 2 for the derivation) the method of Valeur et al. which involves linear least squares analysis of plots of $A_0/(A-A_0)$ versus the reciprocal of the cation concentration at a given wavelength. The stability constant (K_s) is given by the ratio of intercept/slope.^{110, 111}

Formula 1

$$A_o/(A - A_o) = [\epsilon_L/(\epsilon_{ML} - \epsilon_L)][1/K_s[M]] + 1]$$

A_o = free ligand $[M]$ = concentration of cations

A = complexed ligand ϵ_L = molar extinction coefficient of the ligand

K_s = stability constant ϵ_{ML} = molar extinction coefficient of the complex

The determined stability constants for **1** and **2** are given in **Tables 5** and **6**. Stability constants for **1** show near linear plots indicated by the linear correlation coefficients. However, the stability constants for **2** show an increased deviation from linearity on the addition of Mg^{2+} , Na^+ and Li^+ respectively. The reason for this deviation in stability constant for **2** is not known. Furthermore the stability constants for benzo-15-crown-5 were impossible to determine due to the insensitivity of the absorbance bands to the addition of cations. Although the stability constant calculations were disappointing, the values for **1** however do indicate the expected order of selectivity in the cations as Mg^{2+} , Li^+ and Na^+ respectively.

Table 5 Determination of stability constants on the aliquot addition of Mg^{2+} , Na^+ and Li^+ to **1** (at 322nm)^a

Cation	Stability Constant log K_s	Correlation Coefficient
Mg^{2+}	3.8	1.000
Na^+	3.4	0.998
Li^+	3.7	0.987

^a Solutions were 2×10^{-5} M and the stability constant (K_s) is represented as $\log K_s$

Table 6 Determination of stability constants on the aliquot addition of Mg^{2+} , Na^+ and Li^+ to **2** (at 316nm)^a

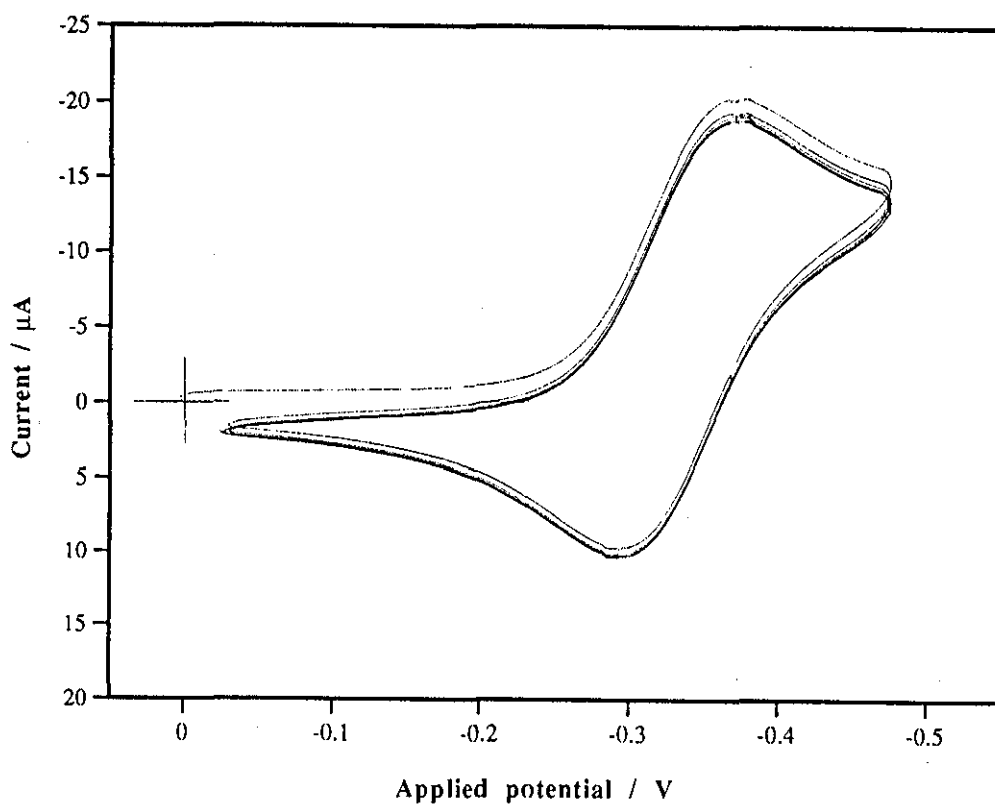
Cation	Stability Constant log K_s	Correlation Coefficient
Mg^{2+}	3.6	0.997
Na^+	3.8	curve
Li^+	-	curve

^a Solutions were 2×10^{-5} M and the stability constant (K_s) is represented as $\log K_s$

5.1.2 Electrochemistry

The diquat dication **5** can be readily reduced, giving rise to a stable radical cation by a one-electron reduction process.^{119, 120} This is shown in **CV 1** which displays the reversible redox process corresponding to the formation of the radical cation ($E^f = -0.33$ V). The highly coloured green radical cation is observed in the vicinity of the electrode/electrolyte interfaces on scanning past this first peak. Extension of the

CV 1 - CVs at 100 mVs^{-1} in an acetonitrile solution containing 0.4 mM of **5** in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode



potential range allows formation of the neutral diradical diquat species. This process initially appears to be reversible ($E^f = -0.82$ V) but on sequential cycling, **CV 2**, the reverse oxidation becomes irreversible. The above cyclic voltammetry experiment was repeated with diquat derivatives **1-4** which in each case showed the reversible formation of the stable radical cation. Formal potentials are given in **Table 7**, conjugation to the electron-donating benzo-15-crown-5 moieties de-stabilising the formation of the radical cation.

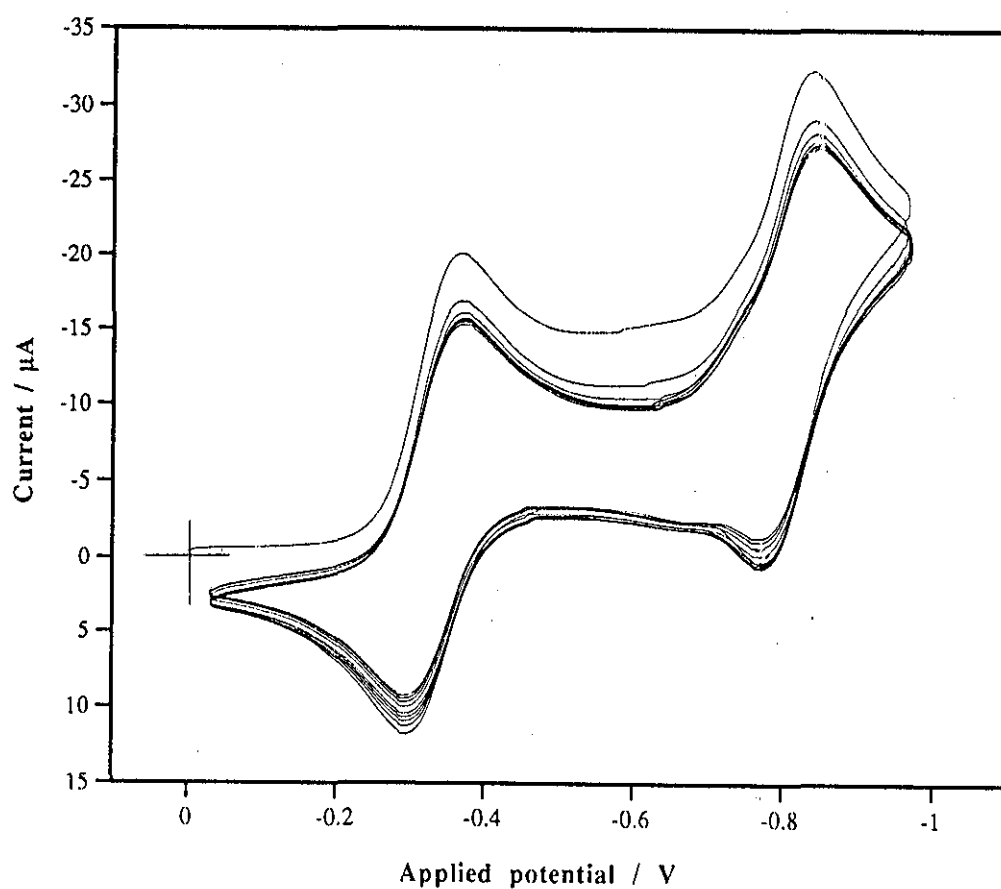
Table 7 Electrochemical data for the diquat and the diquat derivatives **1-5**^a

	$E^f (+2/+1)$
1	-0.41V
2	-0.43V
3	-0.40V
4	-0.43V
5	-0.33V

^a Obtained in acetonitrile solution containing 0.1 M Bu₄NBF₄ as supporting electrolyte. Solutions were 4x10⁻⁴ M in receptor, and potentials were determined with reference to a saturated sodium chloride calomel electrode at 25 ± 2 °C at a scan rate of 100 mVs⁻¹.

However, as with the diquat **5**, sequential scans to the +1/0 peak leads to the redox processes gradually becoming irreversible. This may indicate the formation of a new product possibly dimeric or polymeric in nature. However, a group has observed this

CV 2 - CVs at 100 mVs^{-1} in an acetonitrile solution containing 0.4 mM of **5** in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode



effect with diquat in water and they proposed that it was due to the low solubility of the neutral species.¹²⁰

Complexation studies of Mg^{2+} for **1** and **2**:- It was anticipated that complexation of metal cations would produce anodic shifts in the redox potentials of **1** and **2**. This was investigated with Mg^{2+} , however, addition of excess Mg^{2+} to **1** and **2** resulted in the disappearance of the oxidation peak for the +2/+1 redox process. The reversible redox process +2/+1 which was shown to be stable and reversible in the previous study was probed for the binding of cations. It was found that the addition of an excess of Mg^{2+} cation to the complexes **1** and **2** resulted in the disappearance of the reoxidation wave of the radical cation. This is not totally unexpected as the Mg^{2+} cation on complexation would give the radical cations of **1** and **2** a formal charge of +5 and +3 respectively, which makes it unfeasible to reoxidise these complexes in this potential region. This results in the disappearance of the reduction peak for the +2/+1 redox process due to the exhaustion of the diquat dication due to the prevention of reoxidation.

Electropolymerisation studies using cyclic voltammetry:- One of the aims of the incorporation of the vinyl linkage in the receptors **1-4** is to extend the recognition concept to the preparation of surface modified electrochemical or spectrochemical sensor systems. It was anticipated that electropolymerisation would take place via reductive activation of the vinyl linkages. The diquat derivative 4-vinyl-4'-methyl-N,N'-ethylene-2,2'-bipyridinium has been reported to undergo electropolymerisation by Murray et al.¹²¹ With this in mind the analogous complexes **1-4** were studied. These receptors were studied using cyclic voltammetry at 100mV s^{-1} . Cyclic voltammograms

scanned between 0.00 $\leftrightarrow$ -1.7 V showed a broad irreversible electro-reduction wave at an applied potential more negative than -1.0V and the current decreased with scan number. The electropolymerisation of all the diquat derivatives resulted in the electrogeneration of a non-adherent product and in the case of **1** and **3** a solution colour change. The electrogeneration of non-adherent products rather than polymer films may be due to a number of reasons. In Murray's experiments the presence of dimers or small oligomers (i.e., partly polymerised polymer) greatly facilitates the production of electroreductive film. He noted that highly purified vinyl substituted diquat derivatives polymerise very slowly. The diquat derivatives examined here were shown to very pure which may be one reason for the lack of non-adherent film. Murray also stated that the counter ion in the monomer solution is important in film growth, good film accumulation occurs in ClO_4^- medium, but none in BF_4^- . The supporting electrolyte used in these experiments was Bu_4NBF_4 which is probably another reason why electropolymerisation to form a adherent product does not take place. Also steric factors in **1-4** may also affect the production and rates of formation of adherent films.

5.2 Self-complexation and diquat complexation

The binding of a diquat moiety was first studied by Stoddart et al. who described the complexation in both solution and solid states of the diquat dication, as its bis(hexafluorophosphate) salt, by a range of dibenzo-3n-crown-n ethers ($n=6-12$) and related hosts.¹²² Further to this a series of ^1H NMR experiments conducted by Beer and Rothin showed the binding of a diquat dication by a 2,2'-bipyridyl bis crown ether

ligand which is related to **1** and **6** (Fig. 8).¹²³ From this previous work it seems logical that **1** might form complexes with itself and/or diquat.

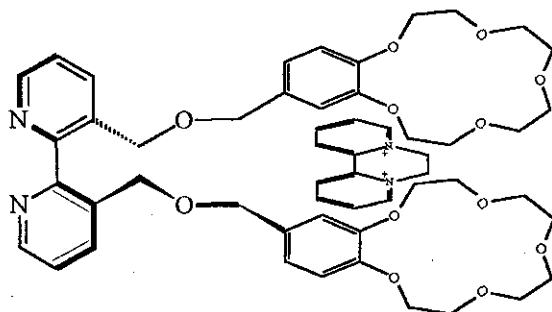


Fig. 8 Complexation of diquat by the 2,2'-bipyridyl bis crown ether ligand

5.2.1 ¹H NMR Spectroscopy

Investigation of self-complexation:- The ¹H NMR spectra were recorded on 1×10^{-2} M solutions of **1** and **2** in deuteriated acetonitrile at $22^\circ\text{C} \pm 2^\circ\text{C}$. The spectrum of **2** was well defined and was interpreted, whereas the spectrum of **1** was broad in nature and it was impossible to resolve at room temperature. However, on increasing the temperature to $70^\circ\text{C} \pm 2^\circ\text{C}$ the spectrum became resolved. This suggests that **1** self associates at room temperature ($22^\circ\text{C} \pm 2^\circ\text{C}$) and only at elevated temperatures is the self complexation broken. Further work has been completed into self complexation by Dr. Oldrich Kocian at Birmingham University. He found that a very dilute (free) solution of **1** gave a different Retardation Factor (R_f) on thin layer chromatography to that of a concentrated (self-complexation) solution. This implies that self-complexation is concentration as well as temperature dependent. Further to this he found that by the

addition of an excess of KPF_6 to a ^1H NMR solution of **1** at room temperature the originally very broad signal became resolved and the spectrum looks the same as the compound at $70^\circ\text{C} \pm 2^\circ\text{C}$. The change in the ^1H NMR spectrum on the addition of K^+ also demonstrates metal cation complexation.

Investigation of diquat complexation:- ^1H NMR spectroscopy was used to study the binding of the diquat dication by **1** and **6** in deuterated acetonitrile. Prior to the addition of the diquat dication the spectrum of **1** showed self complexation. On the 1:1 addition of the diquat dication the spectrum became more resolved. **Tables 8** and **9** reports the significantly large upfield shifts for all the bipyridyl protons of the guest.

Table 8 ^1H NMR spectroscopy chemical shift differences of diquat dication (**5**) guest upon complexation with host **1** in deuterated acetonitrile

	H(6),(6')	H(3),(3')	H(4),(4')	H(5),(5')	($^-\text{NCH}_2$) ₂
Diquat (5)	8.98	8.87	8.81	8.36	5.14
Diquat (5)+ 1	8.78	8.67	8.60	8.15	4.93
$\Delta\delta$	-0.20	-0.20	-0.21	-0.21	-0.21

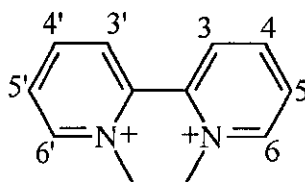


Table 9 ^1H NMR spectroscopy chemical shift differences of diquat dication (**5**) guest upon complexation with host **6** in deuterated acetonitrile

	H(6),(6')	H(3),(3')	H(4),(4')	H(5),(5')	($^+\text{NCH}_2$) ₂
Diquat (5)	8.98	8.87	8.81	8.36	5.14
Diquat (5)+ 6	8.87	8.76	8.69	8.24	5.02
$\Delta\delta$	-0.11	-0.11	-0.12	-0.12	-0.12

The sign and magnitude of these shifts suggest that the planar diquat dication guest intercalates between the two benzo-crown ether subunits of the host, resulting in parallel stacking of the respective aromatic rings **Fig. 9**.

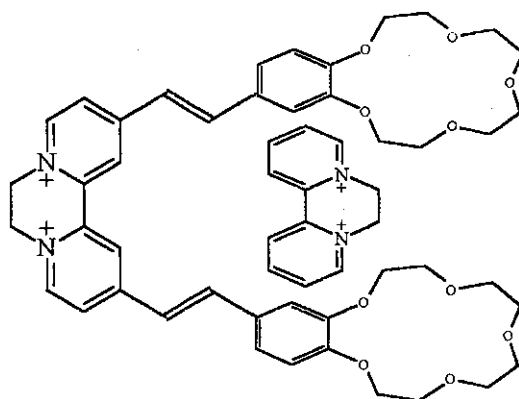


Fig. 9 Diagrammatical representation of the complexation of diquat

5.2.2 UV-vis spectroscopy

In ^1H NMR spectroscopy the diquat derivative **1** complexed **5** by intercalation between the two benzo-15-crown-5 rings. Therefore the effect of the addition of **5** to low concentration (2×10^{-5} M) of **1** and **3** was studied using UV-vis spectroscopy. The

changes on the addition of **5** to **1** and **3** are relatively small which is probably due to the low concentration of the UV-vis solutions (2×10^{-5} M) compared to the ^1H NMR spectroscopy solutions (1×10^{-2} M). This effect has been shown in thin layer chromatography which indicates that complexation does not occur to a great extent at low concentration.

5.3 Anion complexation studies

In the previous chapters an amide group has been solely used in the receptors for anions, where the proton forms hydrogen bonds with the anion such as in Fig. 1. The proton of the vinyl group in the diquat dication receptors **1**, **3** and **6** bear many similarities to the amide protons in Fig. 1. Therefore it is reasonable to propose that the acidic vinyl protons can equally be capable of the complexation of anions in a similar manner to the amide group in Fig. 10. It is also envisaged that the double positive charge on the diquat backbone would enhance anion complexation due to electrostatic interactions. The vinyl groups substituted at the 4,4'-positions of the diquat group produces an acyclic cavity, which is significantly large to provide a cavity that can bind most anions.

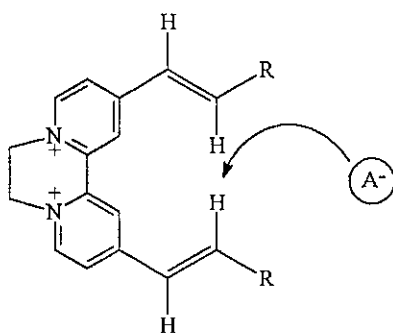


Fig. 10 Proposed site of anion binding as indicated by amide receptor site

5.3.1 UV-vis spectroscopy

UV-vis titrations were performed on **1**, **3** and **6** in acetonitrile with addition of chloride and dihydrogen phosphate anions. Changes in the absorption spectra were noted and found to be related to the quantity of anion present. These absorption changes provided evidence for anion recognition and are detailed in **Tables 10, 11 and 12** where the effects on the absorbance spectrum of the diquat derivatives produce pronounced changes in λ_{max} and absorbances. The peak at 452 nm, 448 nm and 466 nm for **1**, **3** and **6** respectively seem to be the most sensitive to the binding of anions.

The changes in the band at 452 nm for **1** is slightly more pronounced than the band at 448 nm for **3** on the addition of chloride or dihydrogen phosphate. This is probably due to the decreased electron donating ability of benzo-15-crown-5 substituents compared with that of the methoxy substituted phenyl groups.

Table 10 Electronic absorption data for **1** in acetonitrile on the aliquot additions of Cl^- and H_2PO_4^- ^a

$\lambda_{\text{max}}/\text{nm} (10^{-4}\epsilon/\text{M}^{-1} \text{ cm}^{-1})$		
Pure Solvent	H_2PO_4^-	Cl^-
264 (2.47)	284 (3.60)	286 (3.90)
322 (3.24)	328 (3.53)	326 (3.61)
350 (3.43)	*	*
452 (4.52)	458 (2.69)	444 (3.64)

^a Solutions were 2×10^{-5} M. Cl^- and H_2PO_4^- were added as tetrabutylammonium salts to 8×10^{-4} M.

Table 11 Electronic absorption data for **3** in acetonitrile on the aliquot additions of Cl^- and H_2PO_4^- ^a

$\lambda_{\text{max}}/\text{nm} (10^{-4}\epsilon/\text{M}^{-1} \text{ cm}^{-1})$		
Pure Solvent	H_2PO_4^-	Cl^-
322 (3.05)	328 (3.20)	320 (2.90)
354 (2.99)	*	*
448 (4.31)	458 (2.64)	444 (3.70)

^a Solutions were 2×10^{-5} M. Cl^- and H_2PO_4^- were added as tetrabutylammonium salts to 8×10^{-4} M.

* Merges into the peak at 336 nm

Table 12 UV-vis absorption data for **6** in acetonitrile on the aliquot additions of Cl^- and H_2PO_4^- ^a

$\lambda_{\text{max}}/\text{nm}$ ($10^{-4}\epsilon/\text{M}^{-1}\text{cm}^{-1}$)		
Pure Solvent	H_2PO_4^-	Cl^-
244 (6.15)	248 (6.12)	242 (6.42)
290 (10.61)	294 (9.09)	290 (11.19)
368 (7.39)	368 (6.19)	366 (7.01)
466 (4.19)	476 (4.11)	464 (4.03)

^a Solutions were 2×10^{-5} M. Cl^- and H_2PO_4^- were added as tetrabutylammonium salts to 8×10^{-4} M.

The changes in the λ_{max} on the addition of anions are comparatively small when compared with the changes on the addition of cations.

5.3.2 ^1H NMR Spectroscopy

The ^1H NMR spectroscopy study was conducted to determine the orientation and the binding nature between the host and guest. The solvent deuterated DMSO was used in this study, as self-complexation of **1** was not observed (**Fig. 11**), unlike in acetonitrile (section 6.2.1.). Therefore on addition of tetrabutylammonium chloride to a deuterated DMSO ^1H NMR solutions of **1** resulted in the rapid determination of the binding site of the chloride. The most prominent displacements on addition of a 5 x excess of chloride

were those of the $H^{3,3'}$ -bpy protons and the =CH-bpy protons with downfield shifts of $\Delta\delta=0.179$ and 0.130 ppms respectively (Table 13).

Table 13 Changes in the 1H NMR spectroscopy of $H^{3,3'}$ -bpy and =CH-bpy protons on the addition of Cl^-

	Zero Cl^-	5 x excess Cl^-	$\Delta\delta$ in ppm
$H^{3,3'}$ -bpy	9.141	9.320	0.179
=CH-bpy	8.214	8.344	0.130

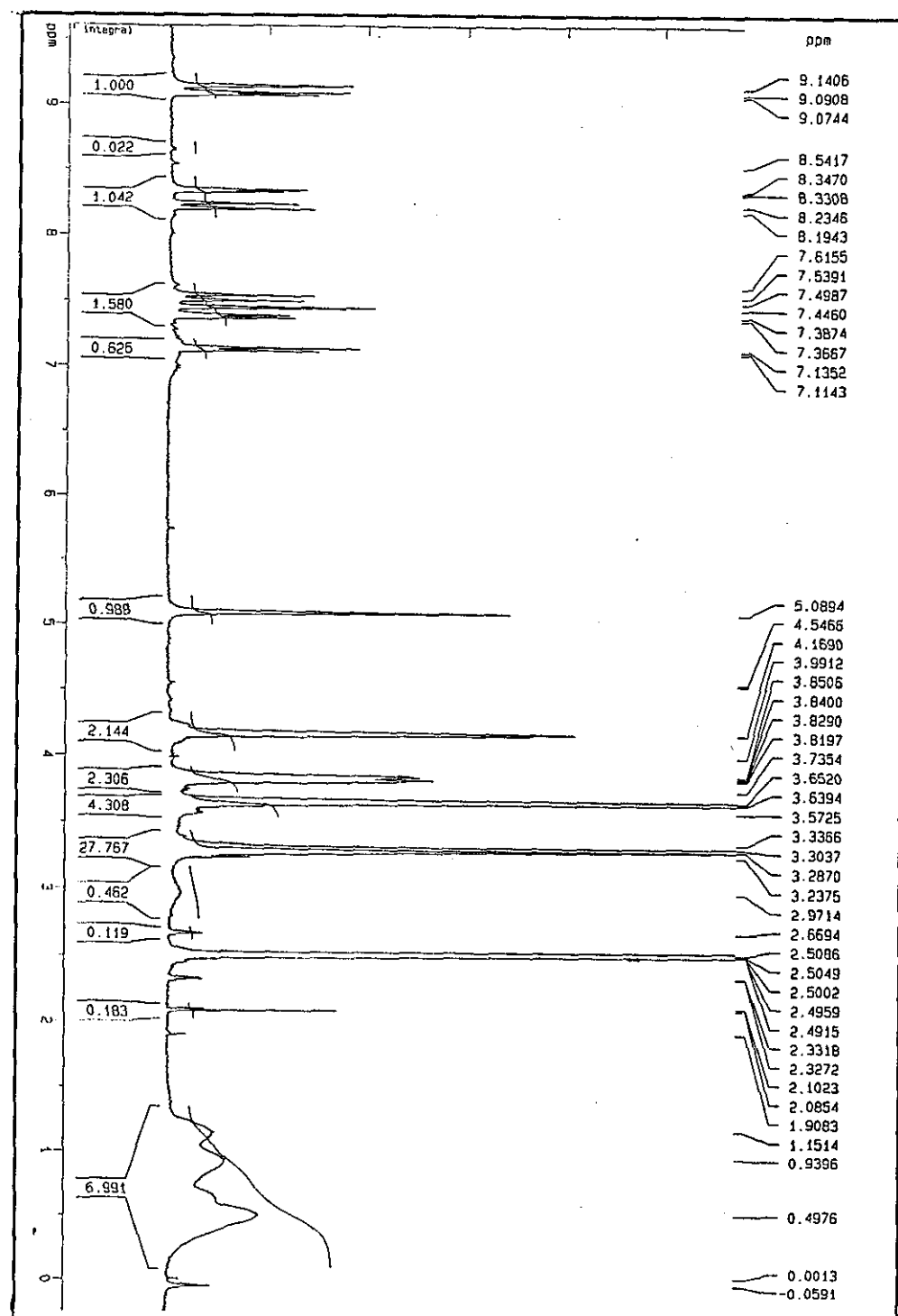
This observation suggests a significant hydrogen bonding interaction with the $H^{3,3'}$ -bpy protons and the =CH-bpy protons which contributes to the overall anion complexation process. It was expected that the prominent displacement would be in the =CH-Ph proton which would give the receptor cavity a similar geometry to that of the amide based receptors. However a number of factors are different between the two cavities:-

1. The =C-H and N -H bond lengths are 1.079 Å and 1.010 Å respectively.¹²⁴

Therefore the cavity created by the =CH-Ph may be too small to accommodate the anion.

2. Steric hindrance caused by the benzo-15-crown-5 groups may prevent the inclusion of the =CH-Ph groups into a cavity.
3. Increased anion binding contribution from the diquat dication due to the closeness of the double positive charge compared to the $[Ru(bipy)_3]^{2+}$

A binding site was therefore proposed, as shown in Fig. 12, unfortunately this cannot be confirmed because the crystal structure has not been determined.

Fig. 11. ^1H NMR of 1 in DMSO

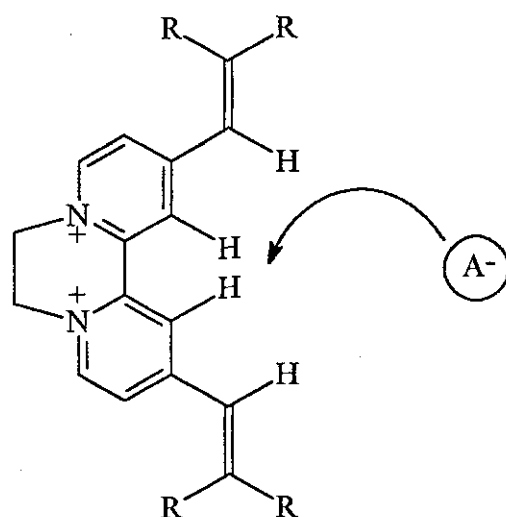


Fig. 12 Proposed anion binding site from ^1H NMR spectroscopy evidence

A receptor which jointly binds both anions and cations is an attractive proposition, as it would provide an alternative to ion exchange resins. Ion exchange is a process which simply swaps the undesirable ions for less troublesome ones. Water softeners, for example take out calcium and magnesium ions and replace them with sodium ions. Recently Reinhoudt et al¹²⁵ have been working on a receptor molecule which can remove salt ions altogether. For example, their receptor molecule can remove both sodium and chloride ions from solution.¹²⁵ As well as the possible application as a water softener it offers opportunities of complexing phosphates, nitrates and radioactive isotopes which should be more effective than using ion exchange resins. With this in mind ^1H NMR spectroscopy was used to look for the joint and possible co-operative binding of anions and cations in complex 1.

The addition of Mg^{2+} prior to the chloride to **1** shows a upfield displacement in the $\text{H}^{3,3'}$ -bpy protons and the =CH-bpy protons of $\Delta\delta=0.033$ and 0.008 ppms respectively (Tables 14 and 15). This indicates that the binding of Mg^{2+} by the benzo-15-crown-5 leads to alterations in the binding properties of the anion receptor sites. This was supported by the moderately small increases in downfield shifts of $\Delta\delta=0.186$ and 0.135 ppms on addition of Cl^- respectively for the $\text{H}^{3,3'}$ -bpy protons and =CH-bpy protons which is approximately a 3.5% change. This result indicates that the joint binding of Cl^- and Mg^{2+} takes place. The increase in downfield shifts on the addition of Cl^- and Mg^{2+} indicates that there may be a small contribution in the binding strength due to co-operative binding.

Table 14 Changes in the ^1H NMR spectroscopy of $\text{H}^{3,3'}$ -bpy protons on the addition of Cl^- and Mg^{2+}

$\text{H}^{3,3'}$ -bpy	Zero Cl^-	5 x excess Cl^-	$\Delta\delta$ in ppm
Zero Mg^{2+}	9.141	9.320	0.179
5 x Mg^{2+}	9.108	9.294	0.186

Table 15 Changes in the ^1H NMR spectroscopy of =CH-bpy protons on the addition of Cl^- and Mg^{2+}

=CH-bpy	Zero Cl^-	5 x excess Cl^-	$\Delta\delta$ in ppm
Zero Mg^{2+}	8.214	8.344	0.130
5 x Mg^{2+}	8.206	8.340	0.135

5.4 Conclusions

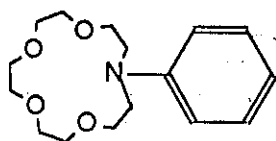
The quaternisation of the bipyridyl ligands to prepare diquat derivatives offers an alternative approach to the use of ruthenium (II) trisbipyridyl derivative as receptors. The linking of the diquat to the benzo-15-crown-5 moiety extends the electronic absorption of the crown ether into the visible region. This is an important development as the extension of the absorption leads to large λ_{max} shifts and colour changes on the addition of Mg^{2+} , Li^+ and Na^+ to **1** and **2**. These effects are not observed with benzo-15-crown-5. The diquat derivative **1** was shown to undergo self-complexation and to bind diquat. ^1H NMR spectroscopy was used to investigate these effects which showed that the diquat dication is intercalated and complexed by the crown ether moieties.

The initial aim for the incorporation of the vinylic group was to perform electropolymerisation to prepare a surface modified electrode. However, this effect was not observed in the supporting electrolyte tetrabutylammonium tetrafluoroborate. The vinylic group bears many similarities to the previously studied amide group (Chapters 3 and 4) which binds anions. ^1H NMR spectroscopy and absorption spectroscopy proved that receptor molecules such as **1** and **6** can bind anion such as Cl^- . The nature of the anion complexation was investigated using ^1H NMR spectroscopy which indicate hydrogen bond formation between the $\text{H}^{3,3'}$ -bpy and $=\text{CH}$ -bpy protons.

Chapter 6 - Electrochemical and spectrochemical studies of N-phenylaza-15-crown-5

6.0 Introduction

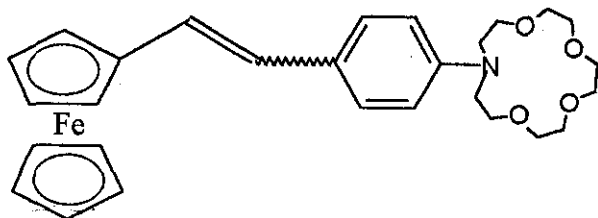
N-Phenylaza-15-crown-5 **1** was originally reported by the renowned supramolecular chemist Vögtle with his colleague Dix.¹²⁶ They found that by the incorporation of a crown ether into a dye molecule, it was possible to directly influence a chromophore on complexation with a cation. The chromophore of this class of receptors showed a shift in λ_{max} on complexation with a number of cations, the magnitude of shift depending on the cation. Influence of the cation on the chromophore is via the nitrogen of the crown ether amine, where the lone pair contribution to resonance is affected to a greater or lesser extent by the positive charge of the guest ion, depending upon the nature of the ion concerned.



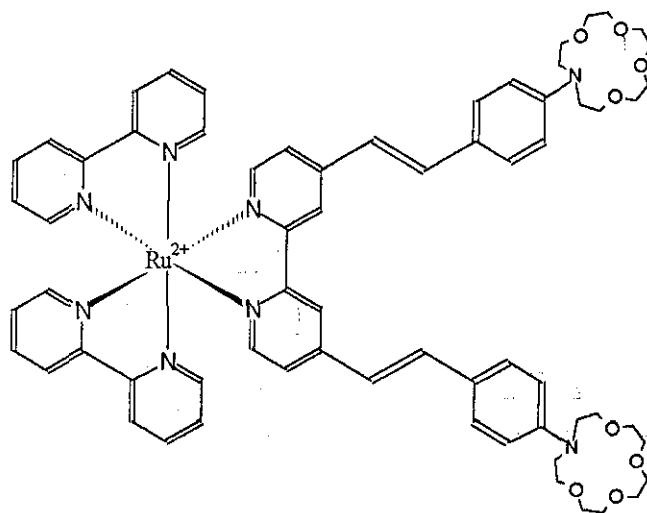
1

A number of groups have also been working on the synthesis of N-phenylaza-15-crown-5 moieties which use redox or fluorophore groups as signalling sites. Beer et al. have utilised the N-phenylaza-15-crown-5 receptor with a number of redox sites including ferrocene **2** and ruthenium(II) polypyridyl complexes such as **3** which electrochemically recognise Mg^{2+} and Na^+ cations.^{65, 97} In the case of **3** the ruthenium(II) polypyridyl unit

also acts as a fluorophore. This results in the spectrochemical recognition of the cations as well.

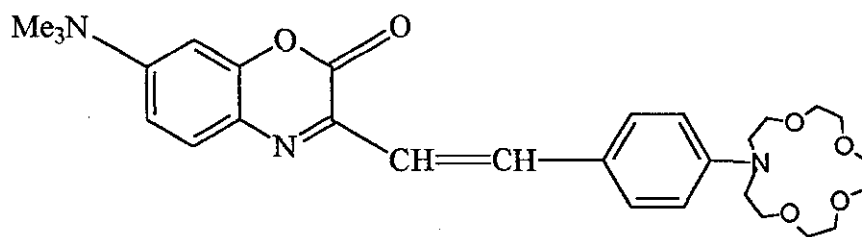


2



3

Valeur and co-workers have been working on the combination of organic fluorescent dyes in combination with crown ethers.^{110, 111} This led to the synthesis of **4** which is monoaza-15-crown-5 linked to a aminobenzoxazino moiety. This links the special photophysical behaviour of aminobenzoxazino with the specific complexing ability of the macrocycle.



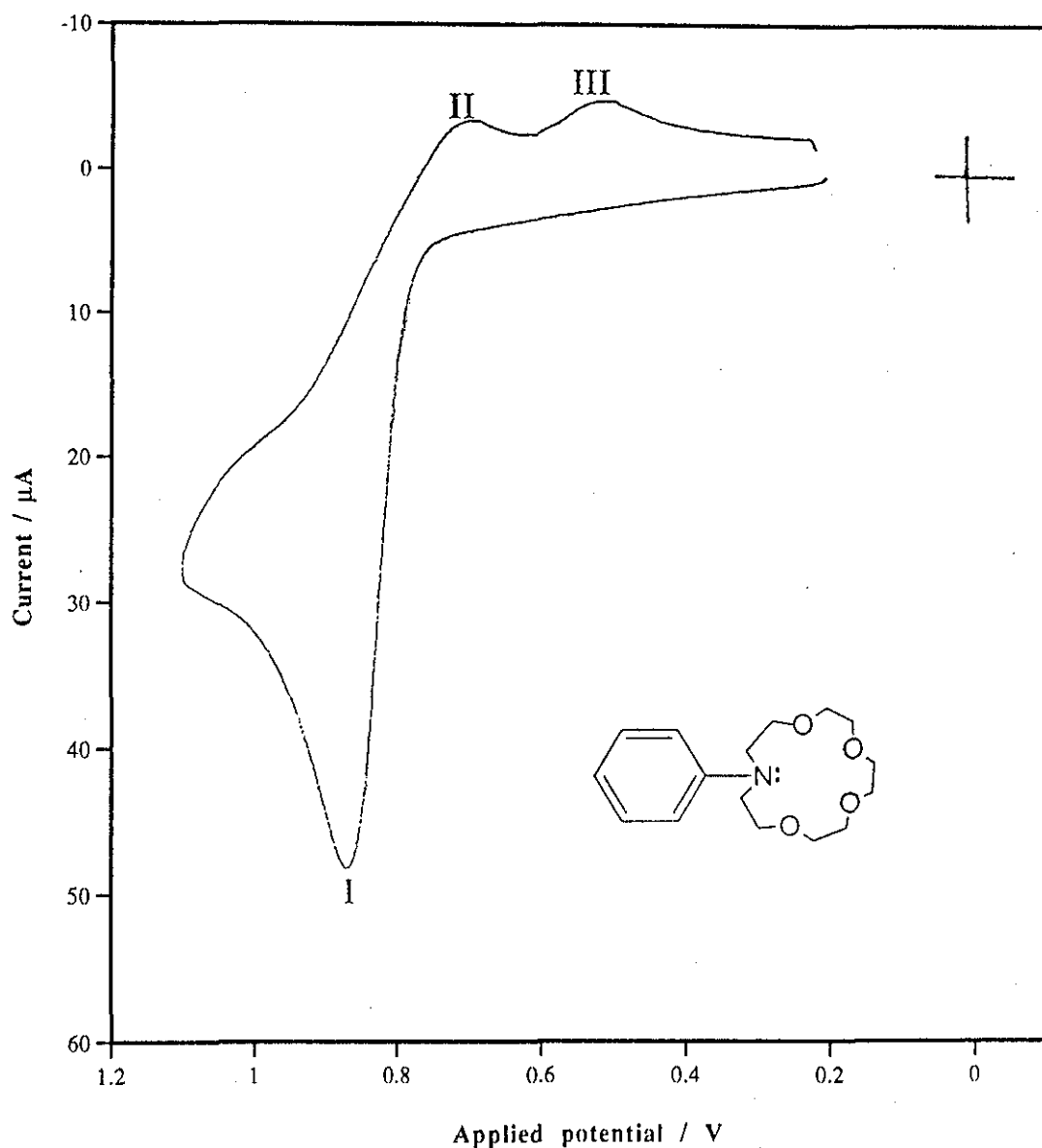
4

In the study of **2** and **3** the electro-oxidation of the crown ether was observed to accompany the electrochemical response of the appended redox centre. This is due to the nitrogen atom of the macrocycle being directly branched onto the phenyl ring. This acts as an electron-donating substituent for the stabilisation of aromatic radical cations. With this in mind it is fair to postulate that a bound cation will influence the lone pair of the nitrogen, and the stability of the aromatic radical cation. This has prompted us to examine the electrochemical oxidation and metal cation-recognition properties of the simple aza-macrocycle N-phenylaza-15-crown-5.

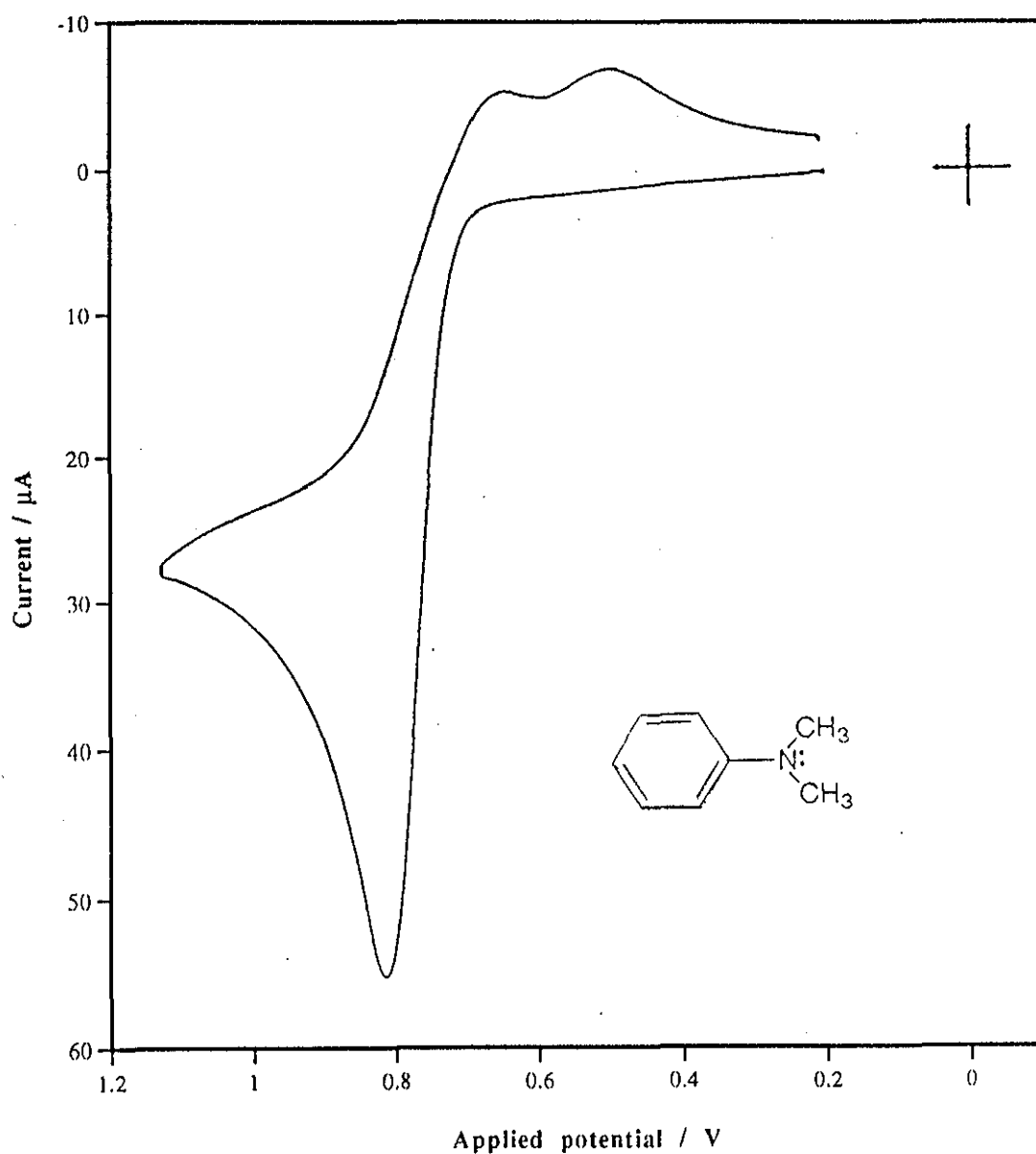
6.1 Cyclic voltammetry of N-phenylaza-15-crown-5

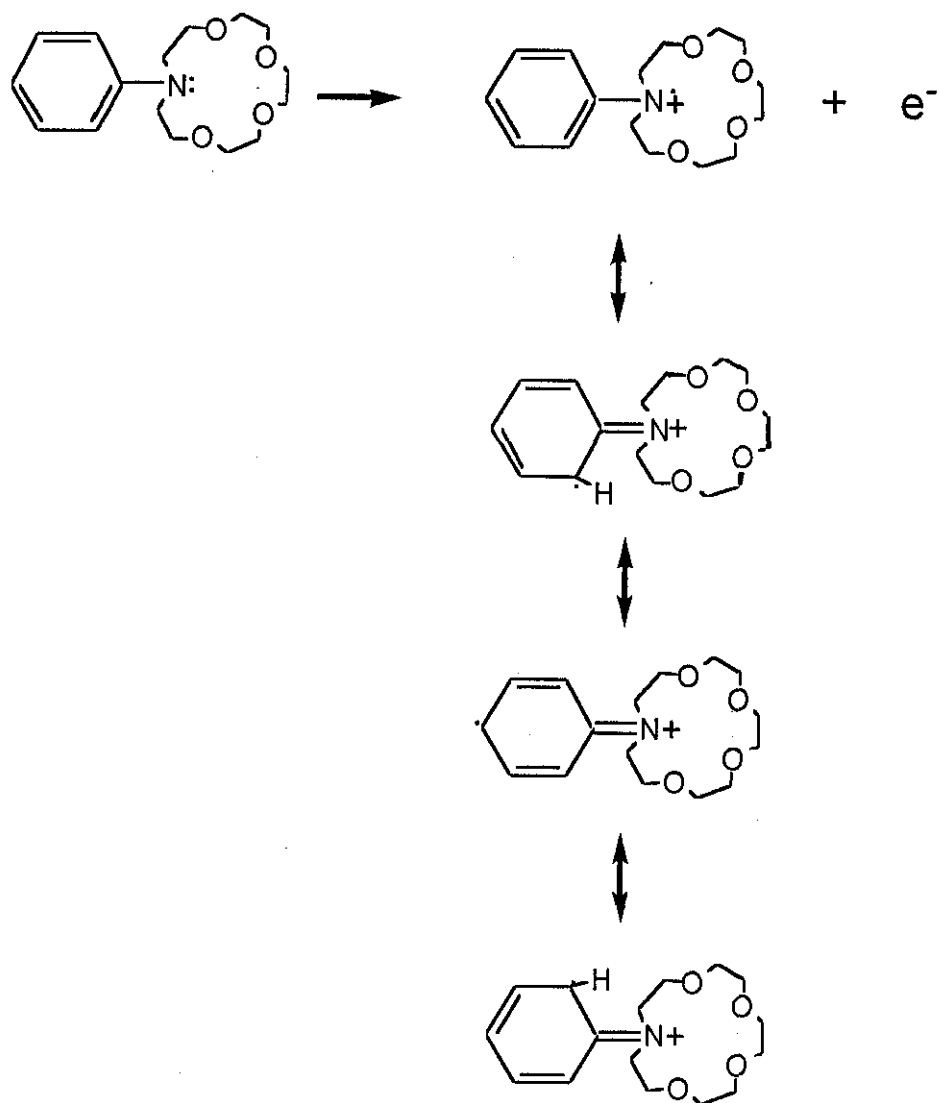
The general form of the cyclic voltammogram shown in **CV 1** for a 1 mM solution of N-phenylaza-15-crown-5 represents an EC_2EE system and paralleled that reported for N,N-dimethylaniline in previous studies^{127, 128} and reproduced here (**CV 2**). Thus the CV of N-phenylaza-15-crown-5 shows a single irreversible peak I ($E_{pa} = 0.84$ V at 50 $mV s^{-1}$) on the first positive scan to give the resonance-stabilised N-phenylaza-15-crown-5 radical cation (**Scheme 1**). A positive shift in E_{pa} from that for N,N-dimethylaniline ($E_{pa} = 0.79$ V at 50 $mV s^{-1}$) is noted, indicating that the aza-15-crown-5 substituent is less effective at stabilising the radical cation than dimethylamine. As

CV1 - CV at 50 mV s^{-1} in an acetonitrile solution containing 1 mM N-phenylaza-15-crown-5 in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode.



CV2 - CV at 50 mV s^{-1} in an acetonitrile solution containing 1 mM N,N -dimethylaniline in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode.

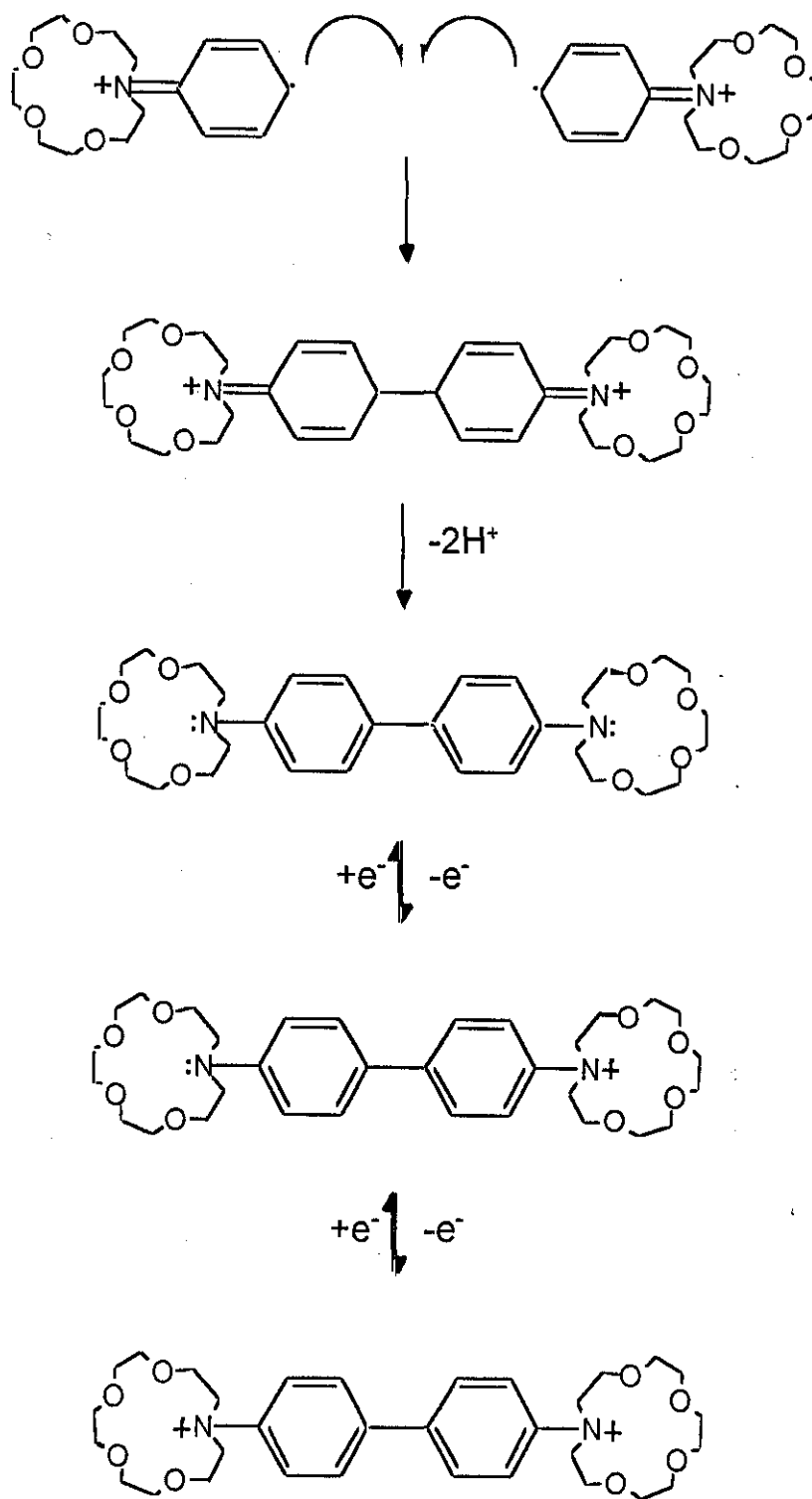




Scheme 1

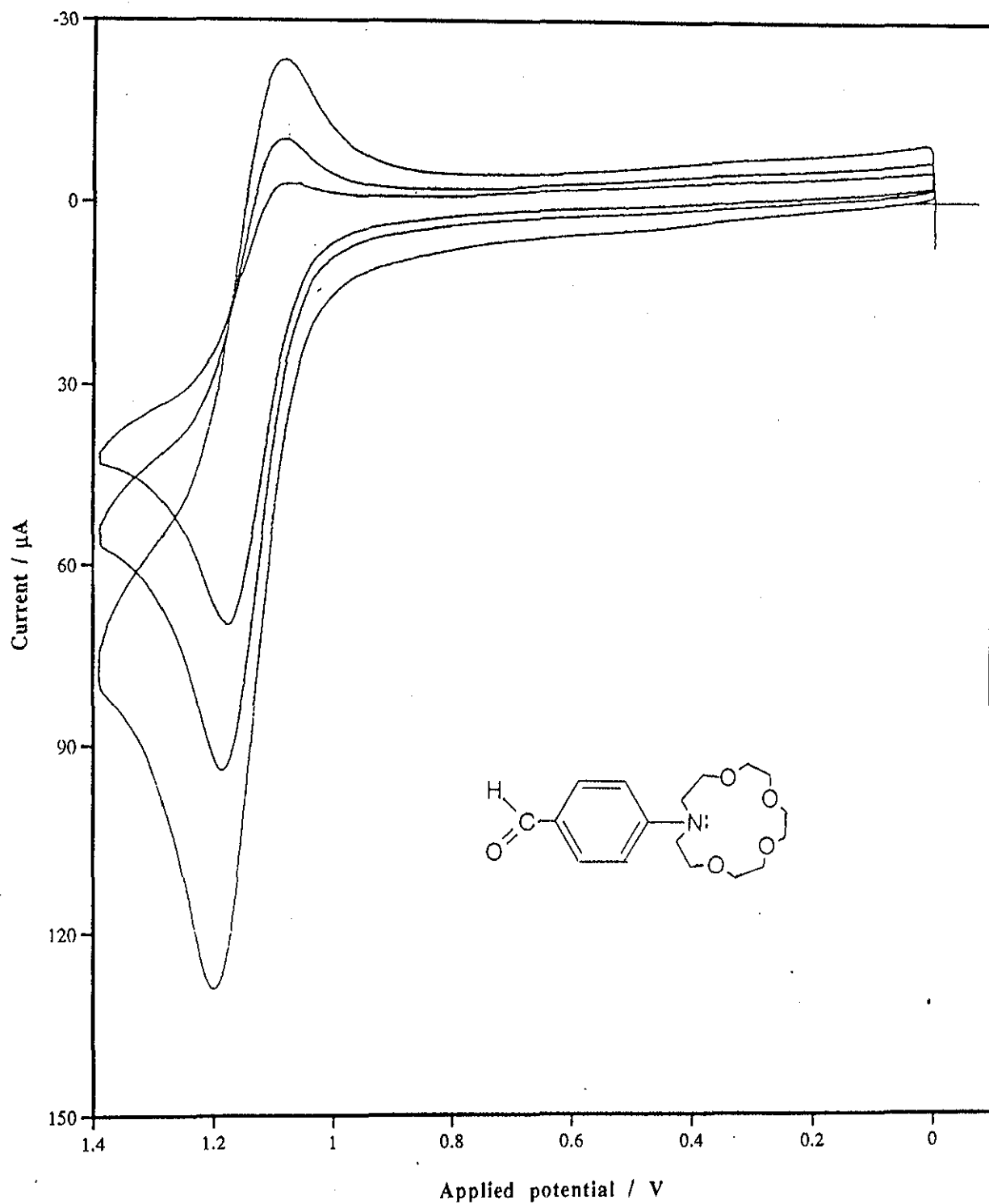
expected for an irreversible process, there is a marked anodic shift in E_{pa} with increasing scan rate (for N-phenylaza-15-crown-5, $E_{pa} = 0.86$ and 0.88 V at 100 and 200 mV s^{-1} respectively).

By analogy with the mechanism for N,N-dimethylaniline,^{127, 128} on generation of the N-phenylaza-15-crown-5 radical cation, a second-order radical cation-radical cation coupling reaction will take place to form (predominantly) the least sterically hindered p-p (C-C) coupled N-phenylaza-15-crown-5 dimer (**Scheme 2**). As for N,N,N',N'-tetramethylbenzidine (TMB), the dimer will be more easily oxidised than the parent monomer and will lose two further electrons at the same potential to form the quinoidal dication (**Scheme 2**). Peaks II and III in the negative scan (**CV 1**) then represent the two one-electron reduction steps of the N-phenylaza-15-crown-5 dimer quinoidal dication. A second positive scan revealed the reversibility of these steps (formal potentials, $E_f = 0.57$ and 0.70 V). That the p-p (C-C) coupled dimer is the main product is suggested by cyclic voltammetry of (4-formyl-benzo)aza-15-crown-5 (**CV 3**) where the 4-formyl substituent prevents coupling at the *para* position. For this molecule, as for the analogous p-dimethylaminobenzaldehyde,¹²⁹ a single quasi-reversible wave is observed with no evidence of redox waves for any o-o coupled dimeric products. Likewise for steric reasons, o-p coupled dimers will be disfavoured for N-phenylaza-15-crown-5. As expected, the formal potentials of the (4-formyl-benzo)aza-15-crown-5 ($E_f = 1.14$ V) and p-dimethylaminobenzaldehyde ($E_f = 1.08$ V, from **Fig. 1** of reference¹²⁹) redox waves are relatively high due to the electron-withdrawing nature of the 4-formyl substituent. That the CV response is quasi-reversible rather than reversible is a further feature of the electron-withdrawing nature



Scheme 2

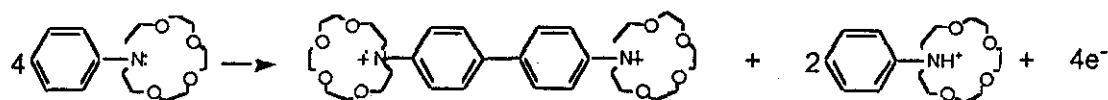
CV3 - CV at 50, 100 and 200 mV s^{-1} in an acetonitrile solution containing 1 mM (4-formyl-benzo)aza-15-crown-5 in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode.



of the 4-formyl substituent. For p-substituted N,N-dimethylanilines, the stability of the radical cation diminishes with an increase in the electron-withdrawing power of the p-substituent.¹²⁸ For the present case, I_p^C/I_p^A increases with scan rate indicating a trend towards reversibility when the radical cation is present at shorter time scales (CV 3).

6.2 Cyclic voltammetry of N-phenylaza-15-crown-5 with addition of pyridine

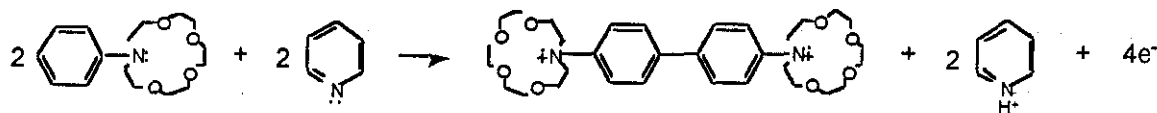
It may be noted that the radical cation-radical cation coupling reaction is associated with de-protonation (Scheme 2). N-phenylaza-15-crown-5 is a moderate base and can serve as a proton acceptor, the protonated form being electroinactive. Hence, as for N,N-dimethylaniline,¹²⁸ the overall reaction will have the following stoichiometry with one electron lost per N-phenylaza-15-crown-5 molecule (Scheme 3).



Scheme 3

As reported for N,N-dimethylaniline,¹²⁸ addition of a stronger base to the electrolyte solution will lead to a two-electron reaction. This was investigated in the present system by addition of pyridine as proton scavenger. On gradual addition of pyridine to a 1 mM solution of N-phenylaza-15-crown-5, I_p^A for peak I increased, with no change in any of the peak potentials. When the concentration of pyridine was equal to that of N-phenylaza-15-crown-5 the current of peak I reached a maximum value of twice that

obtained without the addition of pyridine. Under these conditions, by analogy to N,N-dimethylaniline,¹²⁷ the overall reaction as expressed in **Scheme 4**.



Scheme 4

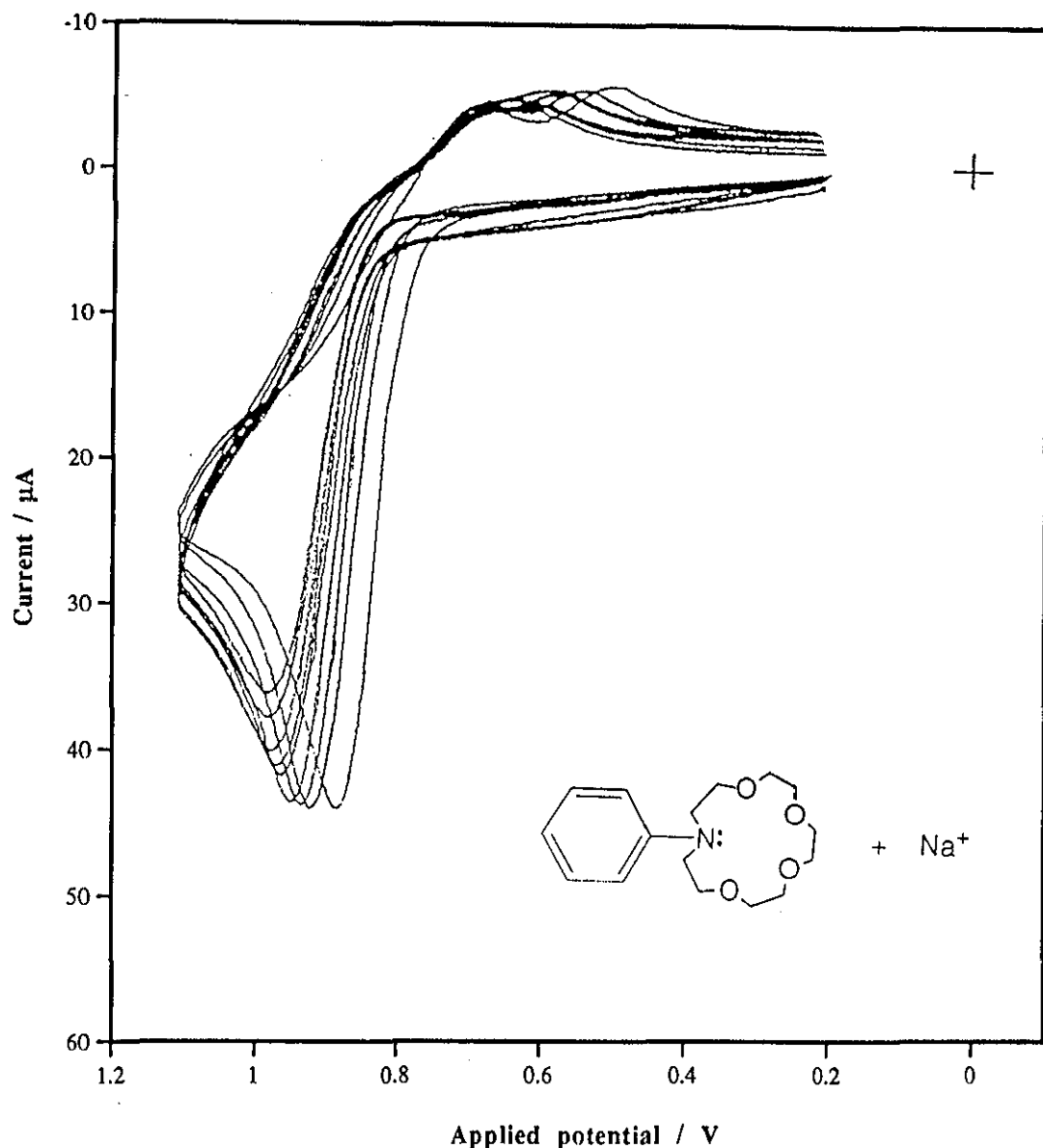
6.3 Electrochemical recognition of metal cations

The metal cation electrochemical recognition properties of N-phenylaza-15-crown-5 and (4-formyl-benzo)aza-15-crown-5 were examined by addition of Na⁺ and Mg²⁺ cations in the form of their perchlorate salts. These cations were chosen for their earlier contrasting effects on the cyclic voltammetric behaviour of N-(4-ferrocenylphenyl)aza crown ethers,^{130, 131} the interpretation being related to their different polarising powers.

Addition of Na⁺ cations.

CV 4 shows the effect on the CVs of titrimetric addition of Na⁺ cations to a 1 mM solution of N-phenylaza-15-crown-5. Macrocyclic binding of Na⁺ cations induces steadily increasing anodic shifts in the potentials of peaks I and III (as labelled in **CV 1**). After addition of 50 times excess of Na⁺ cations no further changes are noted, with peaks II and III being merged. No change in any of the peak potentials were found on addition of Na⁺ cations to a solution of N,N-dimethylaniline, demonstrating in this case the importance of the macrocyclic ring for binding.

CV4 - CV at 50 mV s^{-1} in an acetonitrile solution containing 1 mM N-phenylaza-15-crown-5 in 0.1 M tetrabutylammonium tetrafluoroborate with the addition of sodium perchlorate to 0 (the least positive electro-oxidation wave), 5, 10, 15, 20, 25, 30, 40 and 50 (the most positive electro-oxidation wave) mM. Applied potential versus saturated sodium chloride calomel reference electrode.



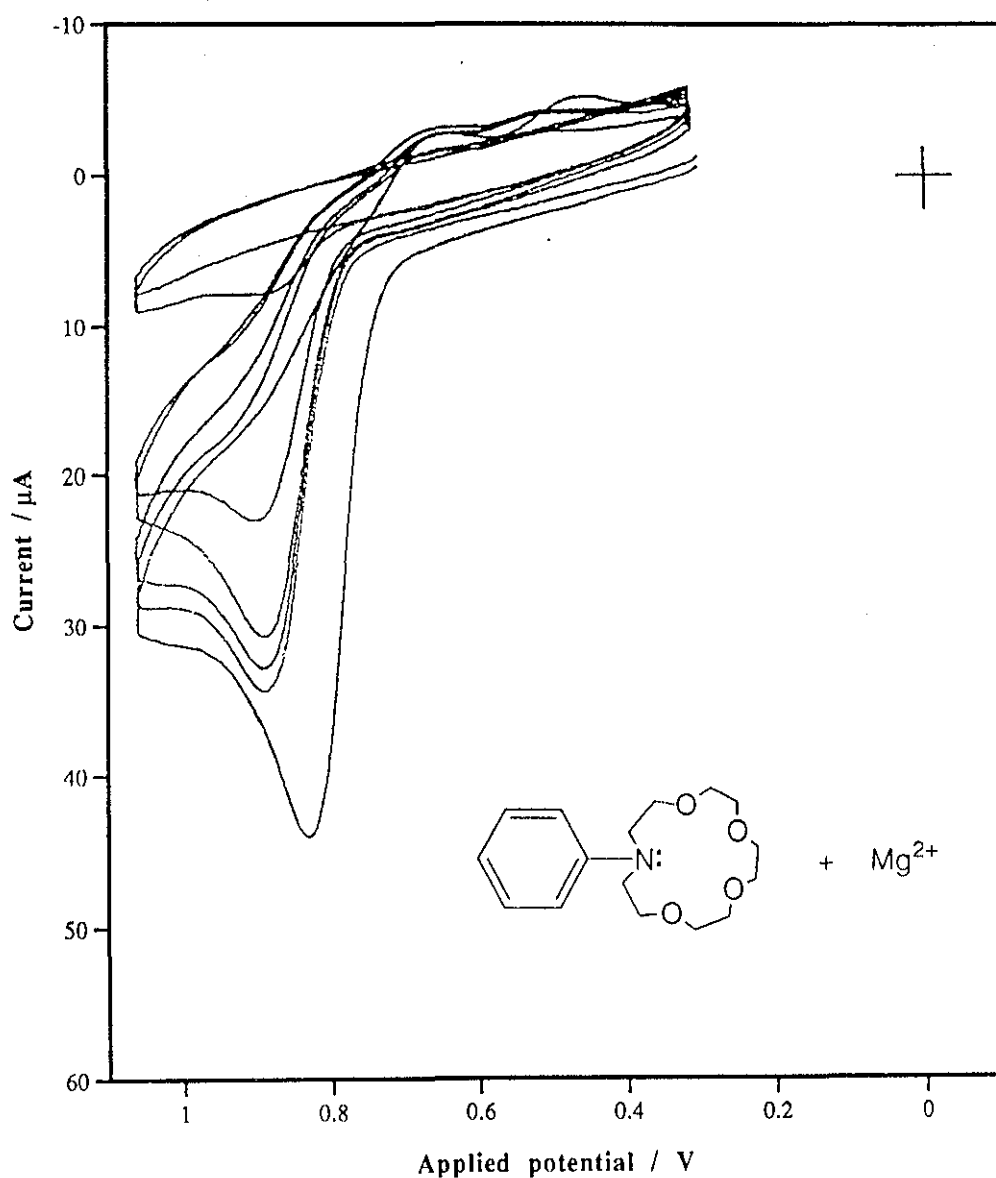
It is of interest to note that the maximum anodic shift ($\Delta E = 110$ mV) observed for peak I is significantly higher than that found for comparable N-(4-ferrocenylphenyl)aza-15-crown-5 electroactive macrocyclic receptor molecules. In the earlier studies on 2, where the electroactive ferrocene reporter group is linked to the N-phenylaza-15-crown-5 by a conjugated olefinic group, the shift was 50 mV for the cis isomer and 65 mV for the trans isomer.¹³¹ Where ferrocene is linked directly to the N-phenylaza-15-crown-5 a shift of 90 mV was reported.¹³² The result reported here shows the benefit of using the phenyl ring electroactivity of the N-phenylaza-15-crown-5 as the reporter, a higher shift than each of these cases being observed. For the N-(4-ferrocenylphenyl)aza-15-crown-5 molecules where ferrocene is linked by a conjugated olefinic group, the N-phenylaza-15-crown-5 irreversible oxidation was unperturbed by addition of Na^+ cations. This was interpreted as being due to fast (on the CV timescale) de-complexation, driven by an increase in the positive charge of the molecule on electro-oxidation of the ferrocene moiety.

Anodic shifts were also noted on monitoring the quasi-reversible redox wave of (4-formyl-benzo)aza-15-crown-5, with a maximum potential shift of 90 mV. For this molecule, the metal cation binding power of the aza-15-crown-5 moiety will be lowered by the presence of the electron-withdrawing 4-formyl substituent resulting in a lower potential shift.

Addition of Mg^{2+} cations.

CV 5 shows the dramatic effect on the CVs of addition of 10 x excess Mg^{2+} cations to a 1 mM solution of N-phenylaza-15-crown-5. The complexation of Mg^{2+} cations is seen

CV5 - A series of seven repetitive CVs at 50 mVs^{-1} in an acetonitrile solution containing 1mM N-phenylaza-15-crown-5 in 0.1 M tetrabutylammonium tetrafluoroborate, with addition of magnesium perchlorate to 10mM after the first CV. Current decrease occurs with scan number. Applied potential versus saturated sodium chloride calomel reference electrode.

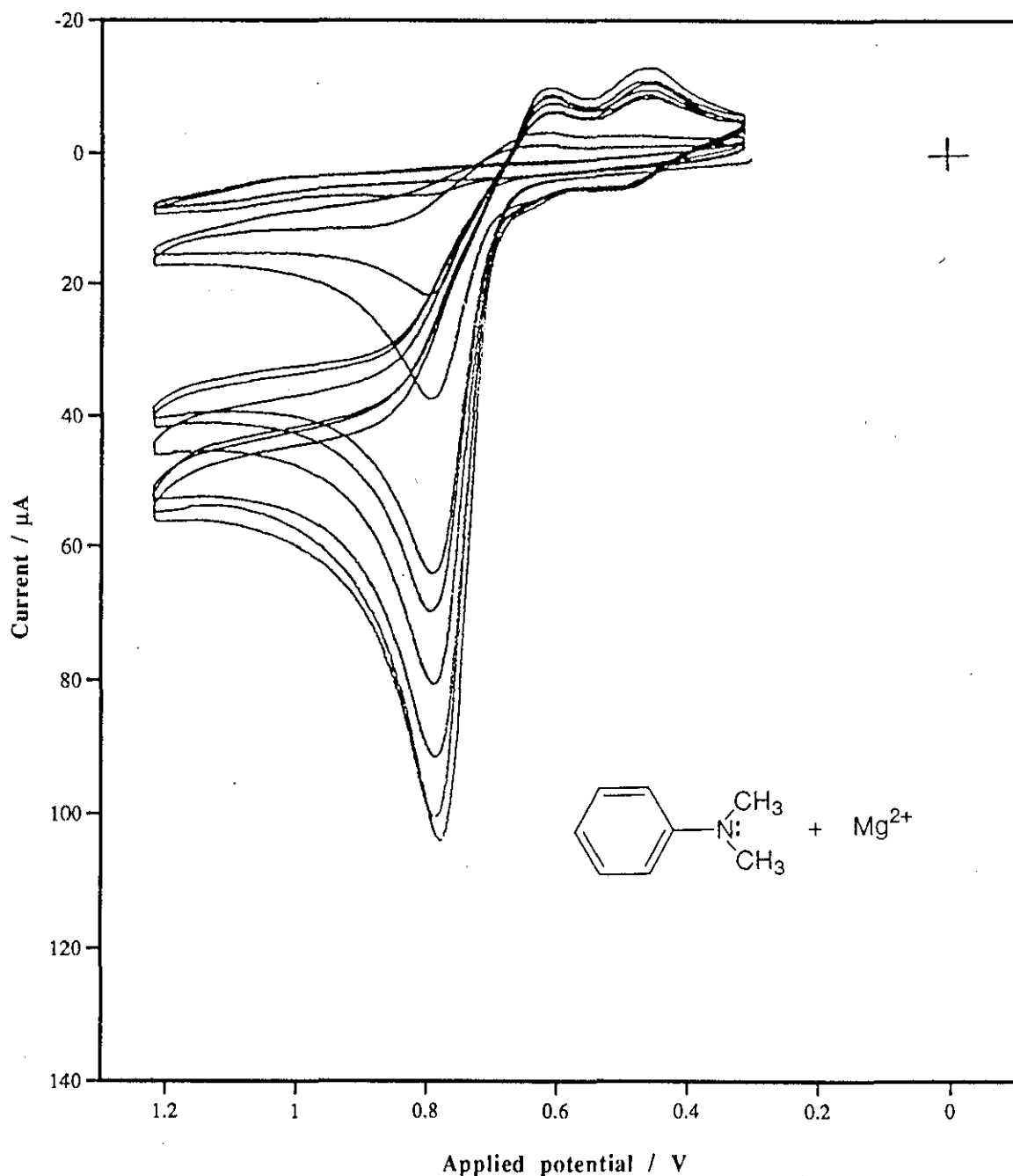


to be slow on the CV timescale. Between scans 1 and 2 an anodic shift ($\Delta E = 60$ mV) is observed, followed by a sequential decrease in the current for peak I. After 210 seconds of repetitive cycling the current has decayed to background levels, oxidation of the complex not being detectable below the solvent oxidation. Earlier studies with *N*-(4-ferrocenylphenyl)aza-15-crown-5 molecules likewise demonstrated the disappearance of the *N*-phenylaza-15-crown-5 irreversible oxidation wave on complexation of Mg^{2+} cations. In the present case, that the observed changes in the CVs (CV 5) represent slow kinetics of complexation, rather than electrode passivation was confirmed by recording the (unperturbed) reversible ferrocene/ferricenium response on transfer of the working electrode to a separate electrolyte solution. Furthermore, electrochemical cleaning and replacement of the working electrode in the original Mg^{2+} cation-containing *N*-phenylaza-15-crown-5 solution gave no recovery of the redox processes.

Whilst no changes in any of the peak potentials were found on addition of Na^+ cations to a solution of *N,N*-dimethylaniline, repetitive CVs (CV 6) demonstrate complexation of Mg^{2+} cations. Whilst this result confirms the importance of the nitrogen atom in the complexation process, it is clearly not exclusively involved in the case of *N*-phenylaza-15-crown-5. Thus for *N,N*-dimethylaniline the current does not decay to background levels until after 360 seconds of repetitive cycling.

Similar effects were noted on monitoring the quasi-reversible redox wave of (4-formylbenzo)aza-15-crown-5, the current decaying to background levels after 220 seconds of

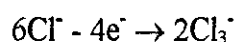
CV6 - A series of eleven CVs at 50 mV s^{-1} in an acetonitrile solution containing 1 mM *N,N*-dimethylaniline in 0.1 M tetrabutylammonium tetrafluoroborate, with addition of magnesium perchlorate to 10 mM after the first CV. Current decrease occurs with scan number. Applied potential versus saturated sodium chloride calomel reference electrode.



repetitive cycling. This indicates that the Mg^{2+} cation complexation rate of the N-phenylaza-15-crown-5 is unaffected by the presence of the 4-formyl substituent.

Addition of Cu^{2+} cations.

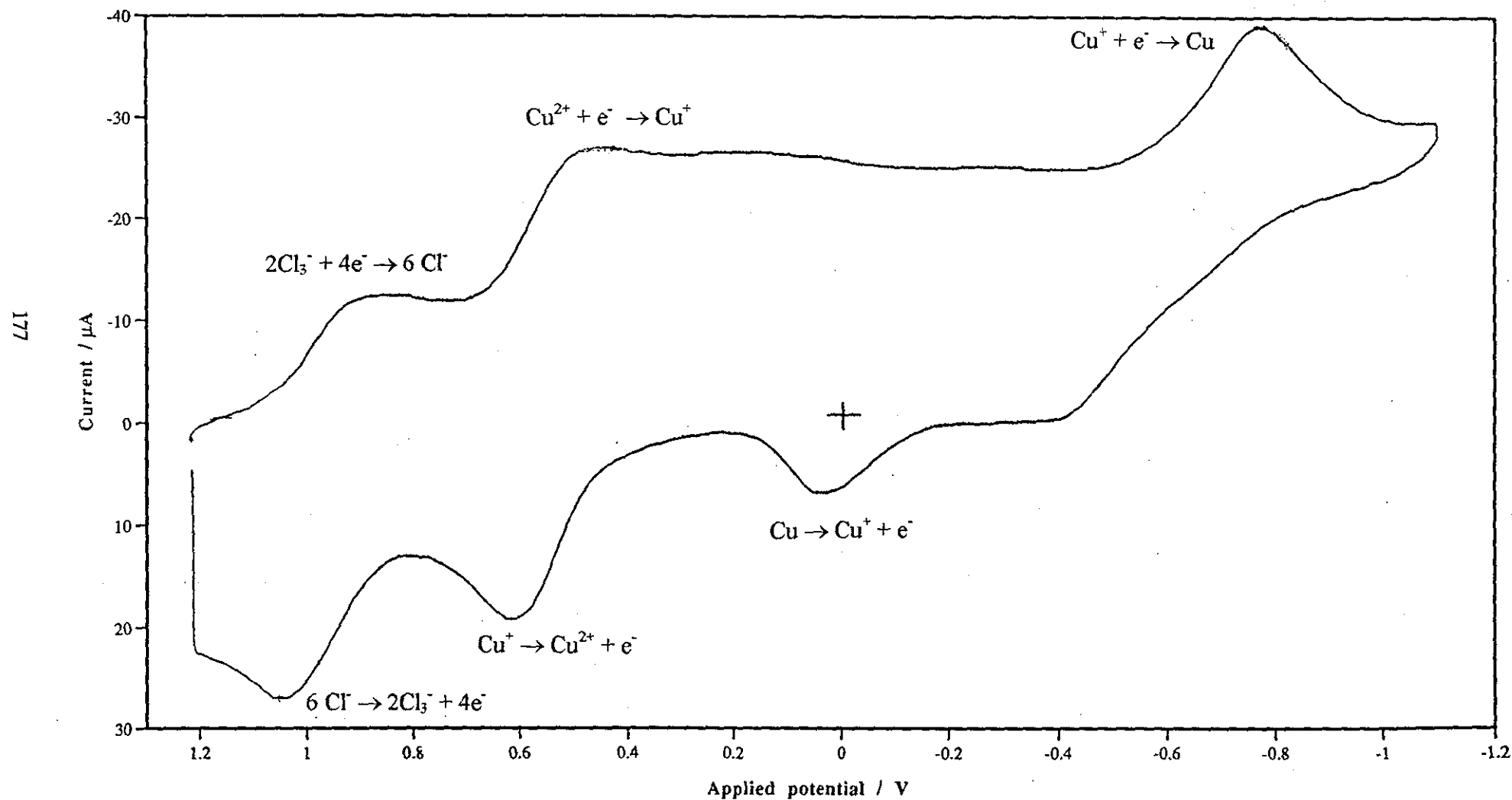
Marji and co-workers demonstrated the complexation of the Cu^{2+} species by N-phenylaza-15-crown-5 in methanol using absorption spectroscopy.¹³⁴ Unlike NaClO_4 and $\text{Mg}(\text{ClO}_4)_2$ which exhibit no electrode reactions in the examined region, CuCl_2 shows a number of electrode processes.¹³⁵ These require interpretation before any complexation studies can take place. In aqueous media the Cu^+ species is unstable and the redox system is Cu^{2+} to Cu. However, with acetonitrile as the solvent the Cu^+ species is stabilised, resulting in reduction waves of equal height. An oxidation reaction of Cl^- is also observed (Scheme 5).



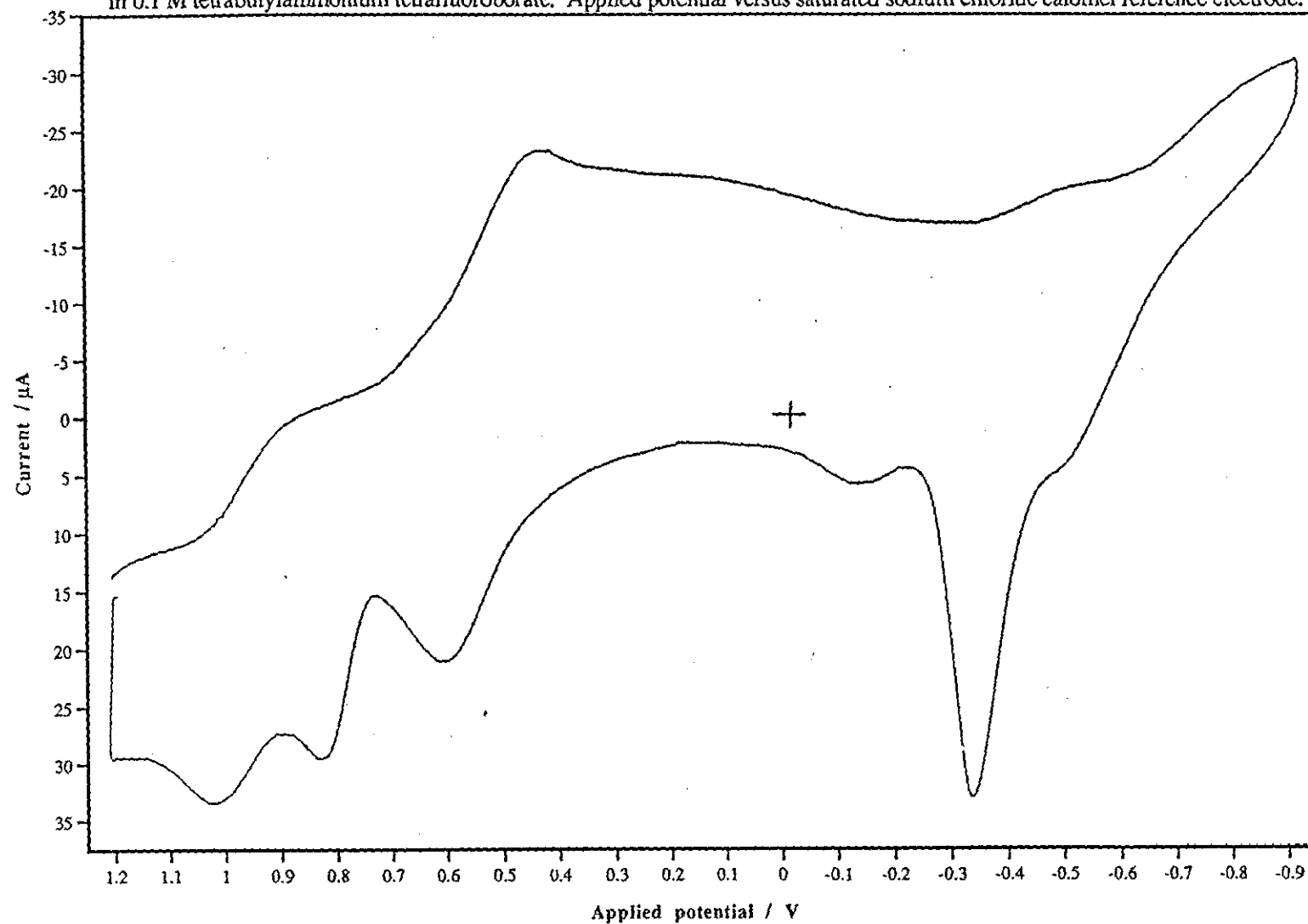
Scheme 5

From literature values the oxidation and reduction peak potentials were assigned to the electrode reactions of CuCl_2 in acetonitrile, shown in CV 7. On the addition of N-phenylaza-15-crown-5 to an equimolar solution of 1mM CuCl_2 in acetonitrile the solution colour changes from yellow to orange, thus indicating the complexation of Cu^{2+} . No shift in peak potentials was observed in oxidation peak of N-phenylaza-15-crown-5. However on the addition of N-phenylaza-15-crown-5 a stripping peak was observed (CV 8). This stripping peak is probably due to the oxidation of the complexed Cu to Cu^+ , the oxidation of the uncomplexed Cu can also be observed and this peak is shifted cathodically. An explanation of this effect may be offered by

CV 7 - CV at 100 mVs^{-1} in an acetonitrile solution containing 1 mM CuCl_2 in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode.



CV 8 - CV at 100 mVs^{-1} in an acetonitrile solution containing 1 mM CuCl_2 and $1 \text{ mM N-phenylaza-15-crown-5}$ in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode.

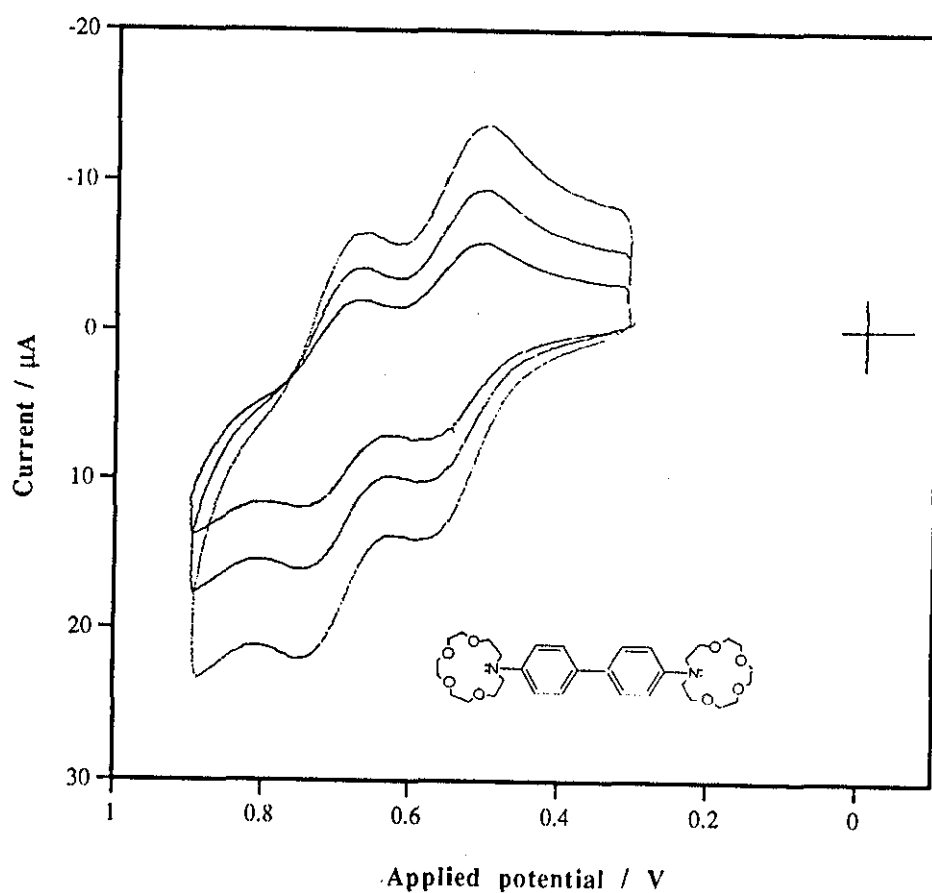


Zhelyazkova who noted that CuCl_2 oxidises the related N,N -dimethylaniline in acetonitrile to give N,N -dimethylaniline radical cation.¹³⁶ He then studied the oxidation reaction using electron spin resonance and electronic spectroscopy, which showed the formation of the $\text{Cu}^+ : N,N\text{-dimethylaniline}^+ : \text{Cl}^-$ complex. This further supports the theory that the stripping peak is due to the complexed Cu.

6.4 Electrogeneration of dimeric products and electrochemical recognition studies

Encouraged by the perturbation on metal cation binding (CVs 4 and 5) of the N -phenylaza-15-crown-5 radical cation/dimer peak III, the electrogeneration and electrochemical recognition properties of the N -phenylaza-15-crown-5 dimer were investigated. The electrogeneration procedure involved constant potential (0.90 V) oxidative electrolysis in the presence of pyridine to give a solution of the N -phenylaza-15-crown-5 dimer dication (Scheme 4), followed by constant potential (0.30 V) reductive electrolysis for conversion to the neutral form. For each step, coulometric measurements verified two-electron stoichiometry and that the electrolyses were exhaustive. Further evidence to confirm that the initial electro-oxidation and subsequent electro-reduction went to completion was obtained by monitoring solution colour changes. On oxidation, the colourless N -phenylaza-15-crown-5 monomer was gradually changed to the bright red N -phenylaza-15-crown-5 dimer cation. Reduction then yielded the olive-green neutral N -phenylaza-15-crown-5 dimer. CV 9a shows the

CV9(a) - CVs at 50, 100 and 200 mV s^{-1} in an acetonitrile solution containing 0.5 mM of electrogenerated N-phenylaza-15-crown-5 dimer in 0.1 M tetrabutylammonium tetrafluoroborate. Applied potential versus saturated sodium chloride calomel reference electrode.



two reversible one-electron redox waves ($E_f = 0.57$ and 0.70 V) to the dimer radical cation and quinoidal dication respectively.

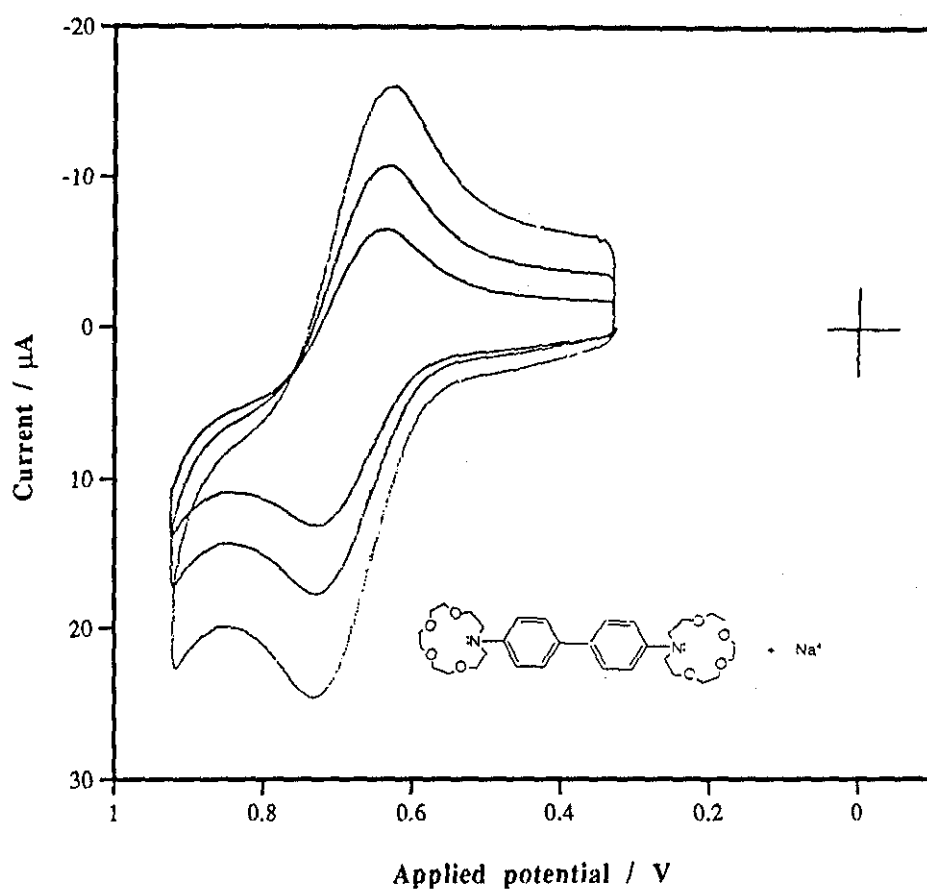
Addition of Na^+ cations.

Titrimetric addition of Na^+ cations to a solution containing 0.5 mM of electrogenerated N-phenylaza-15-crown-5 dimer induced a steady positive shift in the reversible dimer/radical cation redox wave. Changes in the CV response are complete after addition of sodium perchlorate to 50 mM, the two redox waves becoming merged as a single wave (CV 9b). No change in any of the peak potentials were found on addition of Na^+ cations to a solution of electrogenerated N,N-dimethylaniline dimer.

Addition of Mg^{2+} cations.

As for the N-phenylaza-15-crown-5 monomer, addition of Mg^{2+} cations to a solution containing 0.5 mM of electrogenerated N-phenylaza-15-crown-5 dimer produced dramatic effects. Both reversible redox peaks for the dimer disappeared on addition of Mg^{2+} cations. Complexation was confirmed by an olive-green to brown colour change of the solution. As for the N,N-dimethylaniline monomer, similar changes were noted on addition of Mg^{2+} cations to a solution of the electrogenerated N,N-dimethylaniline dimer. However, recurrent CVs for this process again demonstrated slower kinetics of complexation compared to that for the N-phenylaza-15-crown-5 dimer.

CV9(b) - CVs as in **9(a)** after addition of sodium perchlorate to 50 mM.
 Applied potential versus saturated sodium chloride calomel reference electrode.



6.5 Conclusions

These studies have shown that the electrochemical oxidation of the ionophoric macrocycle N-phenylaza-15-crown-5 is sensitive to the binding of metal cations. Addition of Na^+ and Mg^{2+} cations to voltammetric solutions produces significant anodic shifts in the N-phenylaza-15-crown-5 electro-oxidation wave. The charge: radius ratio (polarising power) of the guest cation is of great importance in determining the magnitude of these changes, the Mg^{2+} cations producing the most dramatic effect.

On the addition of CuCl_2 to a N-phenylaza-15-crown-5 solution a colour change was visually observed, which indicates complexation. Cyclic voltammetry was also used to study the addition of Cu^{2+} to N-phenylaza-15-crown-5. A stripping peak was observed for the complexed cation, which is thought to be due to the oxidation of the complexed Cu to Cu^+ .

Constant potential electrolysis of N-phenylaza-15-crown-5 allowed the electrogeneration of solutions containing the p-p coupled N-phenylaza-15-crown-5 dimer. The benefit of this procedure is twofold. Firstly, the dimeric product has two *reversible* one-electron redox waves, the neutral dimer to radical cation process being sensitive to the binding of metal cations. Secondly, the colour changes that take place on cation binding suggest that solutions of electrogenerated dimer could be used for the simple spectrophotometric determination of metal cations.

Finally it should be noted that the investigation presented here would be applicable to many of the numerous aromatic crown ethers that have been described.¹³⁷ In particular, the many monoaza crowns that have been prepared with a para-substituted phenyl on the nitrogen would be amenable to study.^{126, 138} Where the para substituent is hydrogen, the above electrogeneration technique would produce the corresponding dimer with likely analogous properties to that of the N-phenylaza-15-crown-5 dimer. Where the para-substituent has electron-donating properties, electro-oxidation is likely to be reversible and particularly applicable to the study of electrochemical recognition.

Chapter 7 - Development of an Optical Chemical Sensor

7.0 Introduction

This chapter presents an opportunity to discuss developments towards a chemical sensor utilising the novel receptor molecules examined in Chapter 3 and 4. The novel receptor molecules in sections 3.1.2 and 4.1.2 have demonstrated that fluorescence emission spectroscopy measurements have been a useful technique in probing the binding of anions and cations. It was with this in mind that work has been undertaken to develop a sensor which utilises the extremely sensitive technique of fluorescence spectroscopy.¹³⁹ This technique is particularly suited for optical sensing since the wavelength of fluorescence radiation detected is different to that of the incident radiation. Also at very low analyte concentrations, the response of an optical fluorescence sensor is linear according to the equation.

$$I_F = KI_O\Phi_F\epsilon lC$$

I_F = intensity of measured fluorescence C = concentration of reagent

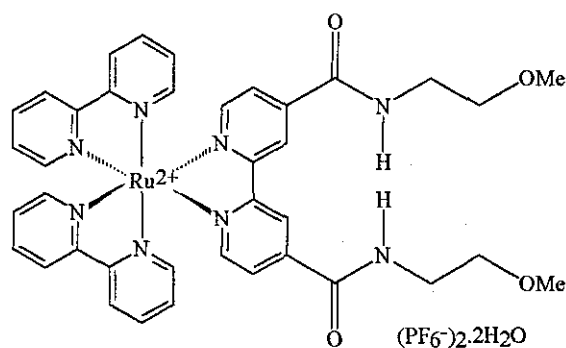
I_O = intensity of incident light ϵ = molar absorptivity

Φ_F = quantum yield of fluorescence l = optical pathlength

K = constant related to instrument and sensor configuration

The basic instrumentation associated with optical sensors is relatively simple, consisting of a light source, a photodetector, monochromator or filters. The development of optical sensors has been accelerated by the use of fibre optics as wave guides. Fibre

optics have low light losses and can be used for the remote monitoring of distant locations, where several locations can be monitored using a star coupler which divides the light into multiple channels. A number of research groups have been working on the development of fibre optic chemical sensors based on the use of immobilised chemical reagents (reagent phase) interfaced with the optical fibre.¹⁴⁰⁻¹⁴² However, in this study fibre optics have not been used due to the high cost involved. This work has been conducted to prove the principle that our receptor molecules could provide the basis of an optical sensor. The development of sensors incorporating anion and cation receptors (such as in Chapters 3 and 4) may lead to the monitoring of biochemical, chemical, medical and environmental important anions and cations. Possible applications include the selective detection of anions such as chloride, both *in vitro* (eg., excreted urine) and *in vivo* (in plasma and biomembranes). Development of methods for the detection of chloride, the major extracellular anion is important, as chloride is involved in the maintenance of proper water distribution, osmotic pressure and normal anion-cation balance. Low serum chloride values are associated with chronic pyelonephritis, Addison's disease, renal failure and diabetic acidosis, and high serum chloride values with dehydration, congestive heart failure and general kidney pathology.¹⁴³ A number of methods have been used in the determination of chloride such as coulometric, spectrophotometric and potentiometric. With a view towards the advancement of chemical sensor technology we have conducted preliminary research into the development of an optical sensing membrane incorporating **1** for the determination of chloride.



7.1 Instrumentation

The aim of this part of the project was to design a simple battery powered and cost effective optical sensor for Cl^- utilising the fluorescent properties of receptors such as **1**. The addition of anionic guest species may cause an enhancement (Cl^-) or quenching (H_2PO_4^-) effect in the fluorescence of the receptor molecule. This makes the design of a sensor easier as it only needs to sense changes in the intensity rather than variation in the wavelength value. A schematic representation of the sensor is shown in **Fig. 1**.

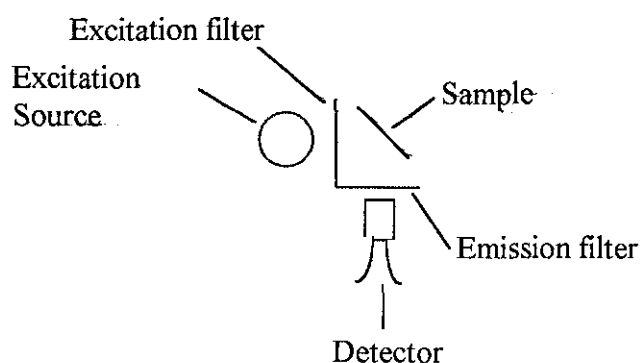


Fig. 1 Diagrammatic representation of the sensor system

The excitation source is a blue light emitting diode (LED) ($\lambda_{\text{max}} \approx 460 \text{ nm}$) which corresponds to the absorbance λ_{max} of the MLCT band. This LED provides a stable and moderately intense light source. Emission from complex **1** is detected using a light dependent resistor (LDR). Optical filters provided the excitation (Kodak, Wratten No. 47) and emission (Kodak, Wratten No. 22) wavelengths shown in Fig. 2.

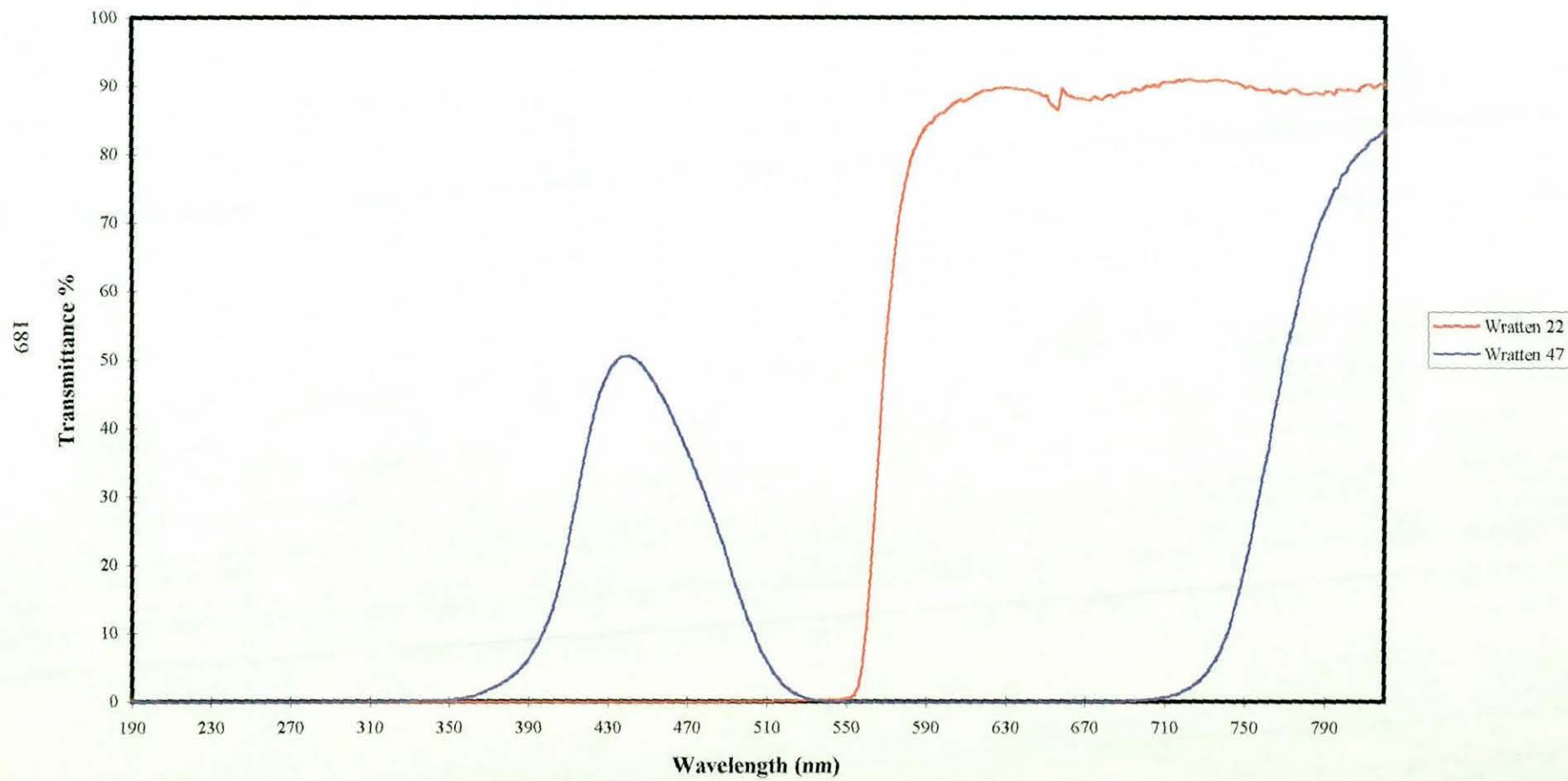
From Fig. 2 it can be seen that there is no overlap of the filters in the visible region, it is therefore reasonable to assume that any change in the intensity signal from the detector is due to fluorescence. The electronic circuit which provides the excitation source is a very simple circuit incorporating an LED, a series resistor and four 1.5 V batteries as the power source (Fig. 3).

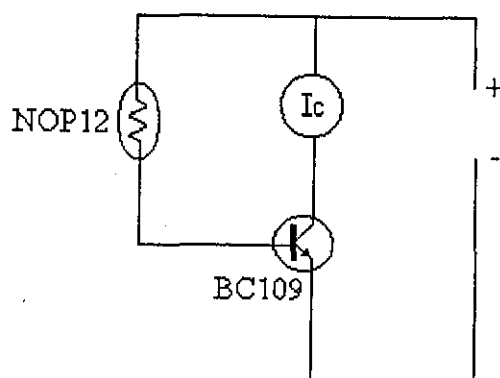


Fig. 3 Circuit diagram of excitation source

Detection of the fluorescence is achieved by the use of a LDR, a transistor is used to amplify the signal, as with the excitation source the power is supplied by four 1.5 V batteries. The resistance of the LDR decreases with an increase in light intensity which leads to an increase in current. The transistor amplifies this signal to provide the current output (I_c). Intensity of the light is directly proportional to the current (I_c) and the amplification is dependent on the gain of the transistor.

Fig. 2 - Transmission spectra of Wratten filters No. 22 and 47





I_c is proportional to the intensity of the light

Fig. 4 Circuit diagram of detector and signal amplifier

7.2 Results and Discussion

Preparation of the membrane:- Complex **1** was immobilised in a high molecular weight PVC matrix (solvent THF) and cast onto an alumina substrate using a specially designed clamp. Unfortunately, this gave a non-homogeneous dispersion of the complex in the polymer matrix. In a more detailed study the dispersion of the complex in the polymer matrix could be improved by changing the polymer type, using plasticizers and alternative methods of casting the polymer film (eg. screen printing and spin coating).

The emission properties of receptor **1** have previously been reported in Chapter 3 which details the effect of the addition of anions. It was found that the addition of Cl^- and H_2PO_4^- to **1** caused enhancement and quenching, respectively in the fluorescence emission. It was therefore decided to limit the study to Cl^- as it is more practical to

monitor an increase in intensity rather than a decrease. Cook and Thomson et al reported that the inclusion of $[\text{Ru}(\text{bipy}_3)]^{2+}$ complexes in polymer matrices such as PVC leads to large increases in quantum yields.¹⁴⁴ Immobilisation increases fluorescence due to the rigid polymer matrix. Fluorescence from the complex was also visually observed, thus further supporting the case for the incorporating of **1** into a polymer matrix. This made the process of aligning the optical components easier (as an optical bench was not available). Large quantum yields on the incorporation in polymer matrices made the utilisation of the complexes more feasible than indicated by the solution work which requires a commercial fluorimeter.

All samples were mounted in the sample holder at a fixed angle of 45° to the excitation source and emission detector. It was found that the excitation and emission filters prevented internal reflection, this was proved by mounting a high polished aluminium disk in the sample holder, which produced a dark current of $1.4 \mu\text{A}$. Once the complex immobilised in PVC was mounted in the removable sample holder, it was not removed until all of the experiments were completed. With detailed engineering this gave reproducible results on the remounting of the sample holder. The PVC immobilised complex **1** was immersed in triply distilled water and dried prior to testing in order to remove any excess complex. The results before and after the addition of Cl^- are detailed in **Table 1**. Addition of the Cl^- anions was made by immersing the sample and sample holder in a saturated solution of NaCl in triply distilled water. As expected quenching of the fluorescence was observed which is due to the polar water molecules solvating the cavity. This solvating water was removed by oven drying the sample and an increase in fluorescence intensity due to the binding of the Cl^- within the cavity was

observed. The fluorescence intensity nearly doubled on the addition of Cl^- which is in-line with the fluorescence experiments performed in aprotic acetonitrile.

Table 1. Fluorescence measurement of **1** immobilised within a PVC matrix on the addition of an excess of chloride.

Conditions	Cl^- ^a	Current μA ^b	Comment
No membrane	None	1.4	Dark current
Immobilised complex - Oven dried	None	23.7	Fluorescence 1
Immobilised complex - Wet sample	NaCl/tdw^c	10.4	Quenching of fluorescence
Immobilised complex - Oven dried	NaCl	44	Fluorescence 1 + Cl^-

^a Addition of Cl^- was made by soaking the membrane in triply distilled water (tdw) saturated with sodium chloride

^b Current is proportional to fluorescence intensity

^c Tdw - triply distilled water

These preliminary experiments demonstrate the possibility of utilising such a fluorescence sensor in the monitoring of the binding of anions and cations by a fluorescence based receptor molecules. The major advantages of such a sensor is that the cost of the development unit was less than £20 and it is battery powered which makes it portable and capable of being used for remote monitoring.

Suggested further improvements

1. The preparation of a reproducible and homogeneous complex/polymer matrix

2. Incorporation of fibre optics

The inclusion of a bifurcated optic fibre which couples the excitation source to the emission detector, as shown in Fig. 5.

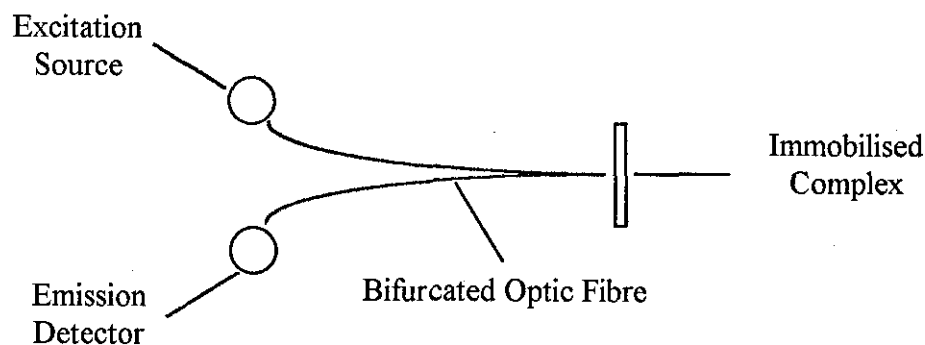
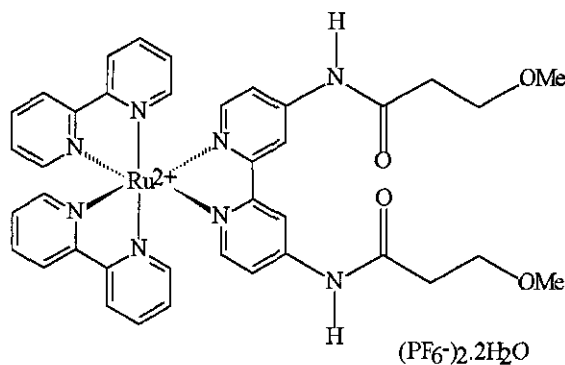


Fig. 5 Coupling of the excitation source to the emission detector

The immobilisation of receptor molecules such as **1** and **2** may lead to the on-line monitoring of sample streams.



Under the experimental conditions in Chapter 3 the fluorescence emission data show the effects detailed in **Table 2** on the addition of Cl^- and H_2PO_4^- to **1** and **2**.

2. Incorporation of fibre optics

The inclusion of a bifurcated optic fibre which couples the excitation source to the emission detector, as shown in **Fig. 5**.

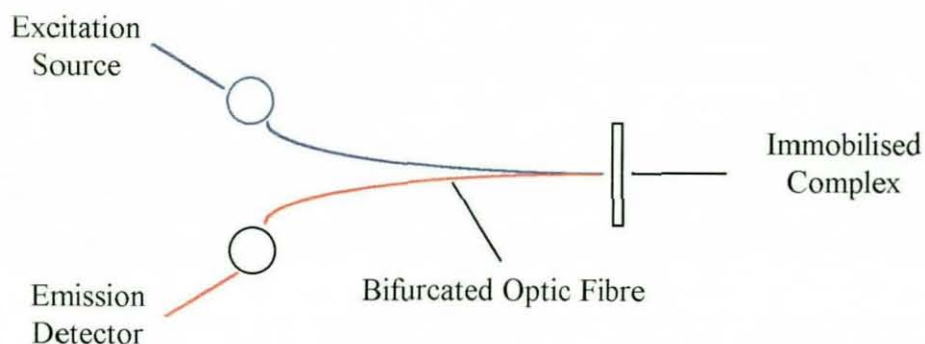
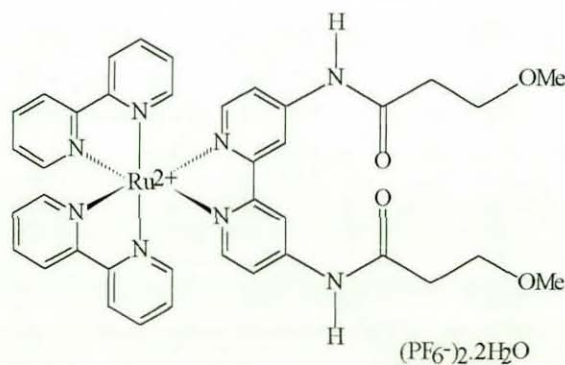


Fig. 5 Coupling of the excitation source to the emission detector

The immobilisation of receptor molecules such as **1** and **2** may lead to the on-line monitoring of sample streams.



2

Under the experimental conditions in Chapter 3 the fluorescence emission data show the effects detailed in **Table 2** on the addition of Cl^- and H_2PO_4^- to **1** and **2**.

Table 2 Changes in emission spectra on the addition of anions to **1** and **2**

	1	2
Cl^-	enhanced	no change
H_2PO_4^-	quenched	quenched

From the results in **Table 2** it is reasonable to assume that the combination of a number of complexes would lead to the selective determination of anions. For example in theory if a sample stream contained a mixture of Cl^- and H_2PO_4^- the concentration of anions can be determined by monitoring the fluorescence of the complexes **1** and **2**. A possible instrument setup is indicated in **Fig. 6**.

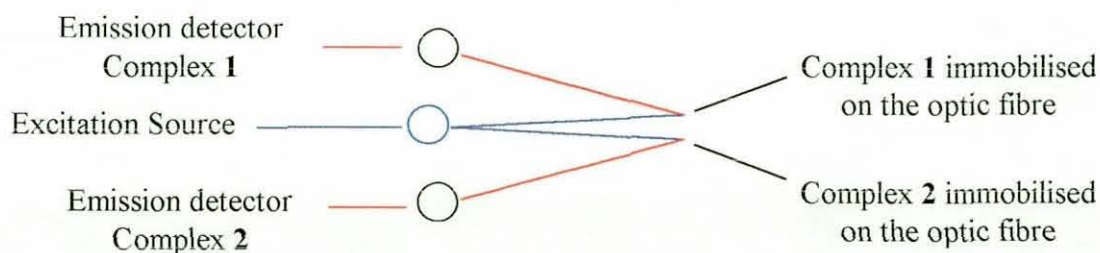


Fig. 6 Possible instrumental setup for the detection of Cl^- and H_2PO_4^- anions

If complex **2** showed a quenching of fluorescence, the presence of H_2PO_4^- (because **2** has been shown to not bind Cl^-) can be established. The presence of Cl^- could be determined using complex **1** by monitoring the change in fluorescence to establish

whether these changes correspond to 100% of H_2PO_4^- . The derivative from the 100% change on the addition of H_2PO_4^- corresponds to the concentration of Cl^- (as Cl^- enhances the fluorescence) on taking into account the selectivity factor.

7.3 Conclusion

The immobilisation of an anion receptor molecule in a polymer matrix has been shown to be sensitive to the binding of anions. More importantly the incorporation of the receptor molecule into a polymer matrix has increased the quantum yield which makes the measurement of the fluorescence simple. This has led to the development of a relatively simple sensor for the detection of fluorescence. Optical sensing of Cl^- anions has been achieved by the development of a battery powered fluorescence detector.

8.0 Summary of Conclusions

The aim of the project was to extend the field of molecular recognition of anions and cations of biochemical, medical, chemical and environmental importance. This has been achieved by the utilisation of receptor molecules including (i) ruthenium(II) trisbipyridyl complexes of acyclic, calix[4]arene and cyclic 2,2'-bipyridine ligands, (ii) macrocycle N-phenylaza-15-crown-5, and (iii) crown ether derivatives of diquat. These receptor molecules have been shown to sense anions and cations, such as chloride, bromide, dihydrogen phosphate, sodium, lithium and magnesium.

The Lewis acidic redox-active and photo-active ruthenium(II) trisbipyridyl moiety in combination with amide CO-NH groups has been successfully incorporated into acyclic, calix[4]arene and cyclic 2,2'-bipyridine ligands. These structural frameworks have produced novel receptors capable of the electrochemical and spectrochemical recognition of anions. Of particular interest is the acyclic receptor **4** (**Chapter 3**) which was shown to be exclusively selective to the binding of H_2PO_4^- anions in preference to Cl^- . The calix[4]arene and cyclic ruthenium(II) trisbipyridyl receptor molecules offered increased structural stability and rigidity of the receptor cavity for H_2PO_4^- and Cl^- with respect to the acyclic derivatives (**Chapter 4**).

With a view to producing spectrochemical and/or electrochemical sensor systems, work has been conducted on the immobilisation of the novel receptor molecules. This has been achieved by the electropolymerisation of vinyl-substituted ruthenium(II) trisbipyridyl complexes or the immobilisation of receptor molecules in a polyvinyl chloride (PVC) matrix (**Chapter 3**). The latter technique has been particularly

successful and has led to the construction of a battery-powered fluorescence detector which has been used for anion sensing (**Chapter 7**).

The quaternisation of the bipyridyl ligands to prepare diquat derivatives (**Chapter 5**) offers an alternative approach to the use of ruthenium (II) trisbipyridyl derivatives as receptors. Synthesis of the diquat derivatives is easier and more cost effective than the preparation of the ruthenium (II) trisbipyridyl derivatives. The linking of the diquat to the benzo-15-crown-5 moiety extends the electronic absorption of the crown ether into the visible region. This was an important development as it led to large colour changes and λ_{max} shifts on the addition of Mg^{2+} , Li^{+} and Na^{+} to the benzo-15-crown-5 derivatives **1** and **2**.

The diquat derivatives (**Chapter 5**) incorporate vinylic groups with the aim of performing electropolymerisation in order to prepare a surface modified electrode. However, this effect was not observed in the supporting electrolyte tetrabutylammonium tetrafluoroborate. The vinylic group bears many similarities to the previously studied amide group (**Chapters 3 and 4**) which binds anions. ^1H NMR and absorption spectroscopy proved that receptor molecules such as **1** and **6** can bind anions such as Cl^{-} . The nature of the anion complexation was investigated using ^1H NMR spectroscopy which indicated hydrogen bond formation between the $\text{H}^{3,3'}$ -bpy and $=\text{CH}$ -bpy protons.

The studies on the ionophoric macrocycle N-phenylaza-15-crown-5 (**Chapter 6**) have shown that the electrochemical oxidation is sensitive to the binding of metal cations.

Addition of Na^+ and Mg^{2+} cations to voltammetric solutions produced significant anodic shifts in the N-phenylaza-15-crown-5 electro-oxidation wave.

Constant potential electrolysis of N-phenylaza-15-crown-5 allowed the electrogeneration of solutions containing the p-p coupled N-phenylaza-15-crown-5 dimer. The benefit of this procedure is twofold. Firstly, the dimeric product has two *reversible* one-electron redox waves, the neutral dimer to radical cation process being sensitive to the binding of metal cations. Secondly, the colour changes that take place on cation binding suggest that solutions of electrogenerated dimer could be used for the simple spectrophotometric determination of metal cations.

References

1. (a) C. J. Pedersen, *J. Am. Chem. Soc.*, 1967, **89**, 7017; (b) C. J. Pedersen, *Aldrichim. Acta*, 1971, **4**, 1.
2. C. J. Pedersen, *Angew. Chem., Int. Ed. Engl.*, 1988, **27**, 1021.
3. (a) J. M. Lehn, *Structure and Bonding*, Springer Verlag, Berlin, 1973, p.1; (b) B. Dietrich, J. M. Lehn and J. P. Sauvage, *Tetrahedron Lett.*, 1969, 2885.
4. J. M. Lehn, *Angew. Chem., Int. Ed. Engl.*, 1988, **27**, 89.
5. D. J. Cram, *Angew. Chem., Int. Ed. Engl.*, 1988, **27**, 1009.
6. (a) F. Vögtle, *Supramolecular Chemistry*, Wiley, Chichester, 1991; (b) G. Gokel, *Crown Ethers and Cryptands*, ed. J. F. Stoddart, The Royal Society of Chemistry, Cambridge, 1991.
7. E. P. Kyba, R. C. Helgeson, K. Madan, G. W. Gokel, T. L. Tarnowski, S. S. Moore and D. J. Cram, *J. Am. Chem. Soc.*, 1977, **99**, 2564.
8. H. K. Frensdorff, *J. Am. Chem. Soc.*, 1971, **93**, 600.
9. (a) R. E. Wolf, and S. R. Cooper, *J. Am. Chem. Soc.*, 1984, **106**, 4646; (b) P. D. Beer, C. J. Jones, J. A. McCleverty and S. Salam, *J. Inclusion Phenom.*, 1987, **5**, 521.
10. S. Shinkai, T. Ogawa, Y. Kusano and O. Manabe, *Top. Curr. Chem.*, 1984, **121**, 67.
11. (a) A. W. Czarnik, *Acc. Chem. Res.*, 1994, **27**, 302. (b) P. D. Beer, O. Kocian, R. J. Mortimer and C. Ridgway, *J. Chem. Soc., Dalton Trans.*, 1993, 2629.
12. J. Rebek Jr., *Science*, 1987, **23**, 1478.
13. A. G. Massey, *Main Group Chemistry*, Ellis Horwood, London, 1990.
14. H. Brockmann and G. Schmidt-Kastner, *Chem. Ber.*, 1955, **88**, 57.
15. T. J. Franklin and G. A. Snow, *Biochemistry of Antimicrobial Action*, Chapman and Hall, London, 1989.
16. C. J. Pedersen, *J. Am. Chem. Soc.*, 1970, **92**, 386.
17. E. Kauffman, J-M. Lehn, and J-P. Sauvage, *Helv. Chim. Acta.*, 1976, **59**, 1099.
18. J. D. Dunitz, M. Dobler, P. Seiler and R. P. Phizackerly, *Acta Crystallogr. Sect. B*, 1974, **30**, 2733; J. D. Dunitz and P. Seiler, *ibid.* 1974, **30**, 2739; M. Dobler, J.

- D. Dunitz and P. Seiler *ibid.* 1974, **30**, 2741; P. Seiler and M. Dobler, J. D. Dunitz. *ibid.* 1974, **30**, 2744; M. Dobler and R. P. Phizackerly, *ibid.* 1974, **30**, 2746.
19. R. Weiss, B. Metz and D. Moras, *Proc. Int. Conf. Coord. Chem. 13th* 1970, **2**, 85.
 20. B. Metz, D. Moras and R. Weiss, *Acta Crystallogr. Sect. B*, 1973, **29**, 85.
 21. D. J. Cram and K. N. Trueblood, *Top. Curr. Chem.*, 1981, **98**, 43.
 22. K. N. Trueblood, C. N. Knobler, E. Maverick, R. C. Helgeson, S. B. Brown and D. J. Cram, *J. Am. Chem. Soc.*, 1981, **103**, 5594.
 23. C. D. Gutsche, Calixarenes, ed. J. F Stoddart, The Royal Society of Chemistry, Cambridge, 1989.
 24. C. D. Gutsche, *Acc. Chem. Res.*, 1983, **16**, 161.
 25. C. D. Gutsche, B. Dhawan, K. H. No and R. Muthukrishnan, *J. Am. Chem. Soc.*, 1981, **103**, 3782.
 26. M. Coruzzi, G. D. Andreetti, V. Bocchi, A. Pochini and R. Ungaro, *J. Chem. Soc., Perkin Trans. 2*, 1982, 1133.
 27. D. Diamond and M. A. McKervery, *Chem. Soc. Rev.*, 1996, **25**, 15.
 28. S. C. Zimmerman, *Top. Curr. Chem.*, 1993, **165**, 72.
 29. C-W. Chen and H. W. Whitlock, Jr., *J. Am. Chem. Soc.*, 1978, **100**, 4921.
 30. J. Rebek Jr., B. Askew, N. Islam, M. Killoran, D. Nemeth and R. Wolak, *J. Am. Chem. Soc.*, 1985, **107**, 6736.
 31. (a) B. Dietrich, *Pure Appl. Chem.*, 1993, **65**, 1457 and references cited therein.
(b) B. Dietrich in Inclusion Compounds vol. 2 (Eds. J. L. Atwood, J. E. D. Davies and D. D. MacNicol) Academic Press, 1984, p.337.
 32. C. S. G. Phillips and R. J. P. Williams, Inorganic Chemistry, Oxford University Press, 1965, p.159.
 33. F. A. Cotton, E. E. Hazen Jr. and M. J. Legg, *Proc. Natl. Acad. Sci., USA*, 1979, **76**, 2551.
 34. P. W. Tucker, E. E. Hazen Jr. and F. A. Cotton, *Mol. Cell. Biochem.*, 1979, **23**, 67.
 35. L. Radom, *Aust. J. Chem.*, 1976, **29**, 1635.

36. S. Goldman and R. G. Bates, *J. Am. Chem. Soc.*, 1972, **94**, 1476.
37. P. D. Beer in *Chemical Sensors*, ed. T. E. Edmonds, Blackie, London, 1987, p.50.
38. C. H. Park and H. E. Simmons, *J. Am. Chem. Soc.*, 1968, **90**, 2431.
39. E. Graf and J-M. Lehn, *J. Am. Chem. Soc.*, 1976, **98**, 6403.
40. B. Metz, J. M. Rosalky and R. Weiss, *J. Chem. Soc., Chem. Commun.*, 1976, 533.
41. F. P. Schmidtchen, *Angew. Chem., Int. Ed. Engl.*, 1977, **16**, 720.
42. J-M. Lehn, E. Soneaux and A. K. Willard, *J. Am. Chem. Soc.*, 1978, **100**, 4914.
43. J-M. Lehn, R. Meric, J-P. Vigneron, I. Bkouche-Waksman and C. Pascard, *J. Chem. Soc., Chem. Commun.*, 1991, 62.
44. T. N. Margulis and L. J. Zompa, *Acta Crystallogr.*, 1981, **B37**, 1426.
45. J. Cullinane, R. I. Gelb, T. N. Margulis and L. J. Zompa, *J. Am. Chem. Soc.*, 1982, **104**, 3048.
46. R. I. Gelb, B. T. Lee and L. J. Zompa, *J. Am. Chem. Soc.*, 1985, **107**, 909.
47. R. I. Gelb, L. M. Schwartz and L. J. Zompa, *Inorg. Chem.*, 1986, **25**, 1527.
48. B. Dietrich, M. W. Hosseini, J-M. Lehn and R. B. Sessions, *J. Am. Chem. Soc.*, 1981, **103**, 3041.
49. G. M. Blackburn, G. R. J. Thatcher, M. W. Hosseini and J-M. Lehn, *Tetrahedron Lett.*, 1987, **28**, 2779.
50. E. Kimura, Y. Kuramoto, T. Koihe, H. Fujioka and M. Kodama, *J. Org. Chem.*, 1990, **55**, 42.
51. A. Bianchi, E. Garcia-Espana, S. Manani, M. Micheloni, P. Orioli and P. Paoletti, *J. Chem. Soc., Chem. Commun.*, 1987, 729.
52. F. Peter, M. Gross, M. W. Hosseini, J-M. Lehn and R. B. Sessions, *J. Chem. Soc., Chem. Commun.*, 1981, 1067.
53. H. M. Colquhoun, J. F. Stoddart and D. J. Williams, *Angew. Chem., Int. Ed. Engl.*, 1986, **25**, 487.
54. T. H. Wirth and N. Davidson, *J. Am. Chem. Soc.*, 1964, **86**, 4325.
55. F. A. Cotton, V. W. Day, E. E. Hazen Jr. and S. Larsen, *J. Am. Chem. Soc.*, 1973, **95**, 4834.
56. O. Kennard and J. Walker, *J. Chem. Soc.*, 1963, 5513.

57. M. L. Fink and M. Bodansky, *J. Am. Chem. Soc.*, 1976, **98**, 974.
58. B. Dietrich, T. M. Fyles, J-M. Lehn, L. G. Pease and D. L. Fyles, *J. Chem. Soc., Chem. Commun.*, 1978, 934.
59. (a) A. Echavarren, A. Galán, J-M. Lehn and J. de Mendoza, *J. Am. Chem. Soc.*, 1989, **111**, 4994 (b) A. Galán, E. Pueyo, A. Salmerón, and J. de Mendoza, *Tetrahedron Lett.*, 1991, **32**, 1827.
60. M. T. Blanda and M. Newcomb, *Tetrahedron Lett.*, 1989, **30**, 3501.
61. M. Newcomb, J. H. Horner and M. T. Blanda, *J. Am. Chem. Soc.*, 1987, **109**, 7878.
62. M. T. Reetz, C. M. Niemeyer and K. Harms, *Angew. Chem., Int. Ed. Engl.*, 1991, **30**, 1472.
63. M. E. Jung and H. Xia, *Tetrahedron Lett.*, 1988, **29**, 297.
64. D. M. Rudkevich, W. P. R. V. Stauthamer, W. Verboom, J. F. J. Engbersen, S. Harkema and D. N. Reinhoudt, *J. Am. Chem. Soc.*, 1992, **114**, 9671.
65. P. D. Beer in *Advances in Inorganic Chemistry*, ed. A. G. Sykes, Academic Press, New York, 1992, **39**, 79.
66. H. Stetter and W. Frank, *Angew. Chem., Int. Ed. Engl.*, 1976, **15**, 686.
67. S. Shinkai and O. Manabe, *Top. Curr. Chem.*, 1984, **121**, 67.
68. P. D. Beer, *Chem. Soc. Rev.*, 1989, **18**, 409.
69. A. Warshawsky and N. Kahana, *J. Am. Chem. Soc.*, 1982, **104**, 2663.
70. A. Kaifer, L. Echegoyen, D. A. Custowski, D. M. Crolis and G. W. Gokel, *J. Am. Chem. Soc.*, 1983, **105**, 7168.
71. M. Rosenblum, in *Chemistry of the Iron Group Metallocenes*, Part 1. Wiley Interscience, New York, 1965.
72. T. Saji, *Chem. Lett.*, 1986, 275.
73. M. Sato, H. Watanabe, S. Abine and S. Akabori, *Bull. Chem. Soc. Jpn.*, 1984, **57**, 1929.
74. P. D. Beer, J. E. Nation and S. L. Brown, *J. Organometl. Chem.*, 1989, **377**, C23.
75. C. D. Hall, I. P. Danks and N. W. Sharpe, *J. Organometl. Chem.*, 1990, **390**, 227.

76. I. Bernal, G. M. Reisner, R. A. Bartsch and B. P. Czech, *Organometallics*, 1991, **10**, 2652.
77. B. Czech and A. Ratajczak, *Pol. J. Chem.*, 1980, **54**, 767.
78. M. Sato, S. Tanaka, S. Akabori and Y. Habata, *Bull. Chem. Soc. Jpn.*, 1986, **59**, 1515.
79. M. Sato, K. Suzuki and S. Akabori, *Bull. Chem. Soc. Jpn.*, 1986, **59**, 3611.
80. P. D. Beer, D. B. Crowe and B. Main, *J. Organometl. Chem.*, 1989, **375**, C35.
81. D. J. Cram, *Science*, 1983, **219**, 1177.
82. D. J. Cram, *Angew. Chem., Int. Ed. Engl.*, 1986, **25**, 1039.
83. P. D. Beer, M. G. B. Drew, A. Ibbotson and E. L. Tite, *J. Chem. Soc., Chem. Commun.*, 1988, 1498.
84. P. D. Beer, E. L. Tite, M. G. B. Drew and A. Ibbotson, *J. Chem. Soc., Dalton Trans.*, 1990, 2543.
85. P. D. Beer, *Chem. Commun.*, 1996, 689 and references therein.
86. P. D. Beer and A. D. Keefe, *J. Organometl. Chem.*, 1989, **375**, C40.
87. P. D. Beer, D. Hesek, J. Hodacova and S. E. Stokes, *J. Chem. Soc., Chem. Commun.*, 1992, 270.
88. P. D. Beer, Z. Chen, A. J. Goulden, A. R. Graydon, S. E. Stokes and T. Wear, *J. Chem. Soc., Chem. Commun.*, 1993, 1834.
89. P. D. Beer, Z. Chen, M. G. B. Drew, J. Kingston, M. Ogden and P. Spencer, *J. Chem. Soc., Chem. Commun.*, 1993, 1046.
90. H-G. Lohr, and F. Vögtle, *Acc. Chem. Res.*, 1985, **18**, 65
91. J. M. Larson, and L. R. Sousa, *J. Am. Chem. Soc.*, 1978, **100**, 1943.
92. J. M. Larson, and L. R. Sousa, *J. Am. Chem. Soc.*, 1977, **99**, 307.
93. (a) F. Fagès, J-P Desvergne, H. Bouas-Laurent, P. Marsau, J-M. Lehn, F. Kotzyba-Hibert, A-M. Albrecht-Gary and M. Al-Joubbeh, *J. Am. Chem. Soc.*, 1989, **111**, 8672. (b) J. P. Konopelski, F. Kotzyba-Hibert, J-M. Lehn, J-P Desvergne, F. Fagès, A. Castellan and H. Bouas-Laurent, *J. Chem. Soc., Chem. Commun.*, 1985, 433.
94. I. Aoki, T. Harada, T. Sakaki, Y. Kawahara and S. Shinkai, *J. Chem. Soc., Chem. Commun.*, 1992, 1341.
95. D. H. Vance and A. W. Czarnik, *J. Am. Chem. Soc.*, 1994, **116**, 9397.

96. P. D. Beer, O. Kocian, R. J. Mortimer and C. Ridgway, *J. Chem. Soc., Chem. Commun.*, 1991, 1460.
97. P. D. Beer, O. Kocian, R. J. Mortimer and C. Ridgway, *J. Chem. Soc., Dalton Trans.*, 1993, 2629.
98. J. M. Calvert, R. H. Schmehl, B. P. Sullivan, J. S. Facci, T. J. Meyer and R. W. Murray, *Inorg. Chem.*, 1983, **22**, 2151.
99. D. R. Crow, *Principles and Application of Electrochemistry*, 3rd edition, Chapman and Hall, London, 1988.
100. R. Greef, R. Peat, L. M. Peter, D. Pletcher, J. Robinson, *Instrumental Methods in Electrochemistry*, Ellis Horwood, Chichester, 1985.
101. P. H. Rieger, *Electrochemistry*, Prentice-Hall, Englewood Cliffs, 1987.
102. A. Juris, V. Balzani, F. Barigelletti, S. Campagna, P. Belser and A. von Zelewsky, *Coord. Chem. Rev.*, 1988, **84**, 85.
103. C. A. Parker and W. T. Rees, *Analyst*, 1960, **85**, 587.
104. K. Nakamaru, *Bull. Chem. Soc. Jpn.*, 1982, **55**, 1639 and reference therein.
105. P. D. Beer, R. J. Mortimer, N. R. Stradiotto, F. Szemes and J. S. Weightman, *Anal. Proc.*, 1995, 419.
106. P. D. Beer, R. J. Mortimer, F. Szemes and J. S. Weightman, *Anal. Commun.*, 1996, **33**, 365.
107. F. Szemes, D. Hesek, Z. Chen, S. W. Dent, M. G. B. Drew, A. J. Goulden, A. R. Graydon, A. Grieve, R. J. Mortimer, T. Wear, J. S. Weightman and P. D. Beer, *Inorg. Chem.*, 1996, **35**, 5868.
108. P. D. Beer, O. Kocian, R. J. Mortimer and C. Ridgway, *Analyst*, 1992, **117**, 1247.
109. P. D. Beer, O. Kocian, R. J. Mortimer and C. Ridgway, *J. Chem. Soc., Faraday Trans.*, 1993, **89**, 333.
110. S. Fery-Forgues, M-T Le Bris, J-P Guetté and B. Valeur, *J. Phys. Chem.*, 1988, **92**, 6233.
111. J. Bourson and B. Valeur, *J. Phys. Chem.*, 1989, **93**, 3871.
112. H. D. Abruña, P. Denisevich, M. Umaña, T. J. Meyer and R. W. Murray, *J. Am. Chem. Soc.*, 1981, **103**, 1.
113. J. N. Demas, E. W. Harris and R. P. McBride, *J. Am. Chem. Soc.*, 1977, **99**, 3547.

114. F. Scandola, M. T. Indelli, C. Chiorboli and C. A. Bignozzi, *Top. Curr. Chem.*, 1990, **158**, 75.
115. K. H. Lieser, *Radiochim. Acta*, 1993, **63**, 5.
116. M. Kawasaki, T. Omori and K. Hasegawa, *Radiochim. Acta*, 1993, **63**, 53.
117. P. D. Beer, Z. Chen, A. J. Goulden, A. Grieve, D. Hesek, F. Szemes and T. Wear, *J. Chem. Soc., Chem. Commun.*, 1994, 1269.
118. R. F. Homer and T. E. Tomlinson, *J. Chem. Soc.*, 1960, 2498.
119. W. R. Boon, *Chem. Ind.*, 1965, 782.
120. E. Steckhan and T. Kuwana, *Berichte der Bunsen-Gesellschaft*, 1974, **78**, 253.
121. T. Ikeda, R. Schmehl, P. Denisevich, K. Willman and R. W. Murray, *J. Am. Chem. Soc.*, 1982, **104**, 2683.
122. B. L. Allwood, H. M. Colquhoun, S. M. Doughty, F. H. Kohnke, A. M. Z. Slawin, J. F. Stoddart, D. J. Williams and R. Zarzycki, *J. Chem. Soc., Chem. Commun.*, 1987, 1054.
123. P. D. Beer and A. S. Rothin, *J. Chem. Soc., Chem. Commun.*, 1988, 52.
124. Handbook of Chemistry and Physics, ed. D. R. Lide, CRC, Boca Raton, 1992.
125. J. Scheerder, J. P. M. van Duynhoven, J. F. J. Engbersen and D. N. Reinhoudt, *Angew. Chem. Int., Ed. Engl.*, 1996, **35**, 1090.
126. J. P. Dix and F. Vögtle, *Angew. Chem. Int., Ed. Engl.*, 1978, **17**, 857.
127. R. Hand and R. F. Nelson, *J. Electrochem. Soc.*, 1970, **117**, 1353.
128. H. J. Yang, D. O. Wipf and A. J. Bard, *J. Electroanal. Chem.*, 1992, **331**, 913.
129. M. Melicharek and R. F. Nelson, *J. Electroanal. Chem.*, 1970, **26**, 201.
130. M.P. Andrews, C. Blackburn, J.F. McAleer and V.D. Patel, *J. Chem. Soc., Chem. Commun.*, 1987, 1122.
131. P.D. Beer, C. Blackburn, J.F. McAleer and H. Sikanyika, *Inorg. Chem.*, 1990, **29**, 378.
132. P.D. Beer, H. Sikanyika, C. Blackburn, J.F. McAleer and M.G.B. Drew, *J. Organomet. Chem.*, 1988, **C19**, 356.
133. P.D. Beer, O. Kocian, R.J. Mortimer, C. Ridgway and N.R. Stradiotto, *J. Electroanal. Chem.*, 1996, **408**, 61.
134. D. Marjii and J. Abraham, *Inorg. Chim. Acta*, 1985, **105**, L3.

135. M. Michlmayr and D. T. Sawyer, *J. Electroanal. Chem.*, 1969, **23**, 387.
136. B. G. Zhelyazkova, *Org. Nucl. Chem. Letters*, 1981, **17**, 141.
137. G.W. Gokel and S.H. Korzeniowski, *Macrocyclic Polyether Synthesis*, Springer-Verlag, Berlin, 1982.
138. J. P. Dix and F. Vögtle, *Chem. Ber.*, 1980, **113**, 457.
139. A. L. Harmer and R. Narayanaswamy in *Chemical Sensors* (ed. T. E. Edmonds), Blackie, 1988, 275.
140. O. S. Wolfbeis, B. P. H. Schaffar and E. Kaschnitz, *Analyst*, 1986, **111**, 1331.
141. M. N. Taib and R. Narayanaswamy, *Analyst*, 1995, **120**, 1617.
142. P. C. Hauser and S. S. S. Tan, *Analyst*, 1993, **118**, 991.
143. N. W. Tietz, *Fundamentals of Clinical Chemistry*, Saunders, Philadelphia, 2nd edn., 1976, p. 879.
144. M. J. Cook, A. P. Lewis, G. S. G. McAuliffe, V. Skarda and A. J. Thomson, *J. Chem. Soc., Perkin Trans. II*, 1984, 1293.

Appendix 1

Equation for the determination of stability constant from emission data

Derivation by Valeur et al.^{110, 111}



$$(C_M)_o - C_{ML} \quad (C_L)_o - C_{ML} \quad C_{ML}$$

$$(C_M)_o = \text{concentration of free metal (M)}$$

$$(C_L)_o = \text{concentration of free ligand (L)}$$

$$C_{ML} = \text{concentration of metal-ligand complex (ML)}$$

$$\epsilon_L = \text{molar extinction coefficient of the ligand (L)}$$

$$\epsilon_{ML} = \text{molar extinction coefficient of the complex (ML)}$$

$$I_F^o = \text{initial fluorescence intensity}$$

$$I_F = \text{fluorescence intensity}$$

$$\Phi_L = \text{quantum yield of the ligand}$$

$$\Phi_{ML} = \text{quantum yield of the metal-ligand complex (ML)}$$

$$\text{Stability } K_s = [ML]/([L][M])$$

$$\text{Stability } K_s = C_{ML} / ((C_M)_o - C_{ML})((C_L)_o - C_{ML}) \quad (1)$$

$$\text{now } I_F^o = \epsilon_L \Phi_L (C_L)_o$$

$$\text{and } I_F = \epsilon_L \Phi_L C_L + \epsilon_{ML} C_{ML}$$

$$= \epsilon_L \Phi_L ((C_L)_o - C_{ML}) + \epsilon_{ML} \Phi_{ML} C_{ML}$$

$$= \epsilon_L \Phi_L (I_F^o / \epsilon_L \Phi_L - C_{ML}) + \epsilon_{ML} \Phi_{ML} C_{ML}$$

$$= I_F^o - \epsilon_L \Phi_L C_{ML} + \epsilon_{ML} \Phi_{ML} C_{ML}$$

$$\therefore C_{ML} = (I_F - I_F^o)/(\epsilon_{ML}\Phi_{ML} - \epsilon_L\Phi_L)$$

assume $(C_M)_o$ is in large excess in equation (1) $(C_M)_o - C_{ML} \approx (C_M)_o = [M]$

$$\therefore K_s = (I_F - I_F^o)/(\epsilon_{ML}\Phi_{ML} - \epsilon_L\Phi_L)/[M](I_F^o/\epsilon_L\Phi_L - (I_F - I_F^o)/(\epsilon_{ML}\Phi_{ML} - \epsilon_L\Phi_L))$$

$$1/K_s[M] = ((\epsilon_{ML}\Phi_{ML} - \epsilon_L\Phi_L)/(I_F - I_F^o))(I_F^o/\epsilon_L\Phi_L - (I_F - I_F^o)/(\epsilon_{ML}\Phi_{ML} - \epsilon_L\Phi_L))$$

$$(1/K_s[M]) + 1 = (I_F^o/(I_F - I_F^o))((\epsilon_{ML}\Phi_{ML} - \epsilon_L\Phi_L)/\epsilon_L\Phi_L)$$

$$I_F^o/(I_F - I_F^o) = (\epsilon_L\Phi_L/(\epsilon_{ML}\Phi_{ML} - \epsilon_L\Phi_L))((1/K_s[M]) + 1)$$

Appendix 2

Equation for the determination of stability constant from absorbance data

Derivation by Valeur et al.^{110, 111}



$$(C_M)_o - C_{ML} \quad (C_L)_o - C_{ML} \quad C_{ML}$$

$$(C_M)_o = \text{concentration of free metal (M)}$$

$$(C_L)_o = \text{concentration of free ligand (L)}$$

$$C_{ML} = \text{concentration of metal-ligand complex}$$

$$\epsilon_L = \text{molar extinction coefficient of the ligand (L)}$$

$$\epsilon_{ML} = \text{molar extinction coefficient of the complex (ML)}$$

$$A_o = \text{initial absorbance}$$

$$A = \text{absorbance}$$

$$\text{Stability } K_s = [ML]/([L][M])$$

$$\text{Stability } K_s = C_{ML} / ((C_M)_o - C_{ML})((C_L)_o - C_{ML}) \quad (1)$$

$$\text{now } A_o = \epsilon_L(C_L)_o$$

$$\text{and } A = \epsilon_L C_L + \epsilon_{ML} C_{ML}$$

$$= \epsilon_L((C_L)_o - C_{ML}) + \epsilon_{ML} C_{ML}$$

$$= \epsilon_L(A_o/\epsilon_L - C_{ML}) + \epsilon_{ML} C_{ML}$$

$$= A_o - \epsilon_L C_{ML} + \epsilon_{ML} C_{ML}$$

$$\therefore C_{ML} = (A - A_o)/(\epsilon_{ML} - \epsilon_L)$$

assume $(C_M)_o$ is in large excess $(C_M)_o - C_{ML} \approx (C_M)_o = [M]$

$$\therefore K_s = (A - A_o)/(\epsilon_{ML} - \epsilon_L)/[M](A_o/\epsilon_L - (A - A_o)/(\epsilon_{ML} - \epsilon_L))$$

$$1/K_s[M] = ((\epsilon_{ML} - \epsilon_L)/(A - A_o))(A_o/\epsilon_L - (A - A_o)/(\epsilon_{ML} - \epsilon_L))$$

$$(1/K_s[M]) + 1 = (A_o/(A - A_o))((\epsilon_{ML} - \epsilon_L)/\epsilon_L)$$

$$A_o/(A - A_o) = (\epsilon_L/(\epsilon_{ML} - \epsilon_L))((1/K_s[M]) + 1)$$

Appendix 3

List of papers published

1. P. D. Beer, R. J. Mortimer, N. R. Stradiotto, F. Szemes and J. S. Weightman, *Anal. Proc.*, 1995, **32**, 419.
2. J. S. Weightman, R. J. Mortimer, P. D. Beer and F. Szemes, Abstract No. 1006, The Electrochemical Society Extended Abstracts, Vol. 95, No. 2, 1581, Chicago, IL, October 8-13th 1995.
3. R. J. Mortimer and J. S. Weightman, *J. Electroanal. Chem.*, 1996, in the press.
4. P. D. Beer, R. J. Mortimer, F. Szemes and J. S. Weightman, *Anal. Commun.*, 1996, **33**, 365.
5. F. Szemes, D. Hesek, Z. Chen, S. W. Dent, M. G. B. Drew, A. J. Goulden, A. R. Graydon, A. Grieve, R. J. Mortimer, T. Wear, J. S. Weightman and P. D. Beer, *Inorg. Chem.*, 1996, **35**, 5868.

Appendix 4

Conferences attended and level of participation

1. 185th Electrochemical Society Meeting, San Francisco, CA, 9-14th May 1994.
2. Poster Presentation at the RSC's Analytical Chemistry Conference, R&D Topics, University of Hertfordshire, 10-14th July 1995.
3. Poster Presentation at the Electrochem '94, Conference, Edinburgh University, 12-16th September 1994.
4. Oral Presentation at the East Midlands Electrochemistry Group Meeting, Leicester University, 30th March 1995.
5. National Instruments Data Acquisition Seminar, Leicester, 14th June 1995.
6. Poster Presentation at the RSC's Analytical Chemistry Conference, R&D Topics and SAC Meeting, Hull University, 10-14th July 1995.
7. Oral Presentation at the 188th Electrochemical Society Meeting, Chicago, IL, 8-13th October 1995.
8. East Midlands Electrochemistry Group Meeting, Coventry University, 3rd April 1996.
9. Physics World Exhibition, Telford Exhibition Centre, 24th April 1996.
10. Participant in the RSC's Analytical Division "Initiative for Emerging Professionals in Analytical Science", 1996, Buxton, 26-29th April 1996.
11. DTI's "Think Digital - Microelectronics in Business" Seminar, Manchester, 4th June 1996.
12. Oral Presentation at the RSC's Analytical Chemistry Conference, R&D Topics, Nottingham Trent University, 22-23rd July 1995.

