

AD-A171 338

DEVELOPMENT OF N(2) SENSOR FOR DETERMINATION OF PN(2)
IN BODY TISSUES(U) ETC LABS INC NORWOOD MA
L S ROBBLEE ET AL AUG 86 C-731 ONR-0458(F)

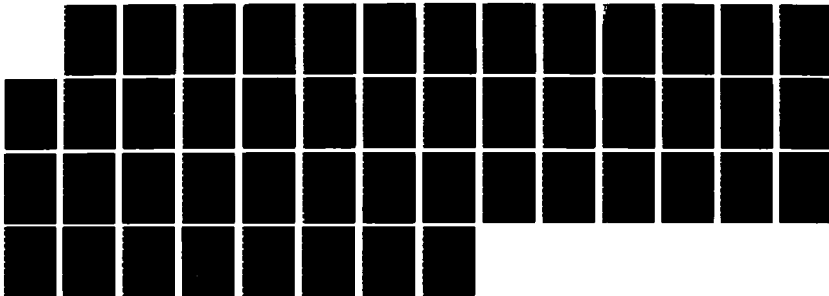
1/1

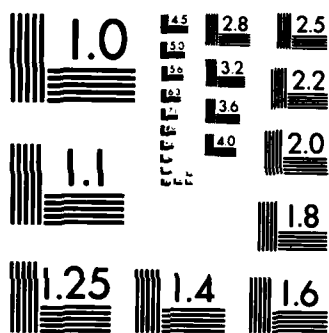
UNCLASSIFIED

N00014-83-C-0458

F/G 7/4

NL





MICROCOPY RESOLUTION TEST CHART
NATIONAL BUREAU OF STANDARDS-1963-A

AD-A171 330

12

Report ONR-0458(F)

DEVELOPMENT OF N₂ SENSOR FOR DETERMINATION OF PN₂ IN BODY TISSUES

CONTRACT N00014-83-C-0458

Principal Investigator: L. S. Robblee
Senior Scientist: T. J. Lewis
Staff Scientists: R. B. Salmonsens
P. A. Huminski
M. S. Blankinship
P. J. Marren

Performance Site: EIC Laboratories, Inc.
111 Downey Street
Norwood, Massachusetts 02062

August, 1986

Final Report for Period 15 June 1983 - 14 June 1986
Approved for Public Release - Distribution Unlimited

Prepared for:

OFFICE OF NAVAL RESEARCH
Department of the Navy
800 North Quincy Street
Arlington, Virginia 22217

436 2-1-86

☆

"Reproduction in whole or in part is permitted
for any purpose of the United States Government"

DTIC FILE COPY

86 8 29 044

Disclaimers

The views and conclusions contained in this document are those of the authors and should not be interpreted as necessarily representing the official policies, either expressed or implied, of the Office of Naval Research or the U.S. Government.

The citation of trade names and names of manufacturers in this report is not to be construed as official government endorsement or approval of commercial products or services referenced herein.

UNCLASSIFIED

SECURITY CLASSIFICATION OF THIS PAGE (When Data Entered)

REPORT DOCUMENTATION PAGE

READ INSTRUCTIONS
BEFORE COMPLETING FORM

1. REPORT NUMBER

2. GOVT ACCESSION NO.

3. RECIPIENT'S CATALOG NUMBER

DA-458(F)

AD171330

4. TITLE (and Subtitle)

5. TYPE OF REPORT & PERIOD COVERED

DEVELOPMENT OF N₂ SENSOR FOR
DETERMINATION OF PN₂ IN BODY TISSUES

FINAL REPORT

15 Jun 83 - 14 Jun 86

6. PERFORMING ORG. REPORT NUMBER

C-731

8. CONTRACT OR GRANT NUMBER(s)

N00014-83-C-0458

9. AUTHOR(s)

Thomas S. Robblee, Thomas J. Lewis, Richard B.
Almosen, Patricia A. Huminski, Michael S.
Frankinship and Peter J. Marren

11. PERFORMING ORGANIZATION NAME AND ADDRESS

3M Laboratories, Inc.
1 Downey Street
Norwood, MA 02062

10. PROGRAM ELEMENT, PROJECT, TASK
AREA & WORK UNIT NUMBERS

NR 201-591

12. CONTROLLING OFFICE NAME AND ADDRESS

Office of Naval Research
100 North Quincy Street
Arlington, VA 22217

12. REPORT DATE

AUGUST 1986

13. NUMBER OF PAGES

46

14. MONITORING AGENCY NAME & ADDRESS (if different from Controlling Office)

15. SECURITY CLASS. (of this report)

UNCLASSIFIED

15a. DECLASSIFICATION/DOWNGRADING
SCHEDULE

16. DISTRIBUTION STATEMENT (of this Report)

Approved for public release; distribution unlimited.

17. DISTRIBUTION STATEMENT (of the abstract entered in Block 20, if different from Report)

18. SUPPLEMENTARY NOTES

19. KEY WORDS (Continue on reverse side if necessary and identify by block number)

Nitrogen sensor, nitrogen electrode, PN₂ measurement, nitrogen tension,
nitrogen complex, ruthenium ammines, ruthenium-dinitrogen complexes,
chemically modified electrodes, ion exchange, Nafion, transition metals,
electrochemical sensor, immobilized ruthenium, heterogeneous catalysis.

20. ABSTRACT (Continue on reverse side if necessary and identify by block number)

Experiments were performed to identify transition metal ion-dinitrogen com-
plex systems whose chemical and electrochemical properties would be favorable
for use in quantitating PN₂, and to develop methods of immobilizing those
metal ions so that the reactions of interest would take place on an electrode
surface. A N₂ sensing system was developed which is comprised of an ion
exchange polymer membrane (Nafion), containing one of the following Ru(III)

FORM 1 JAN 73 1473

EDITION OF 1 NOV 65 IS OBSOLETE
S/N 0102-LF-014-6601

UNCLASSIFIED

SECURITY CLASSIFICATION OF THIS PAGE (When Data Entered)

20. Abstract (Cont.)

cations $[\text{Ru}(\text{NH}_3)_5\text{H}_2\text{O}]^{+3}$, $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+3}$, or $\text{cis-}[\text{Ru}(\text{en})_2(\text{H}_2\text{O})_2]^{+3}$, coated onto a glassy carbon electrode. The electrode is activated for N_2 measurement by applying a reducing potential to generate the N_2 -complexing Ru(II) cation on the electrode surface. The surface Ru(II) cation reacts with N_2 in solution to form a surface Ru(II)- N_2 complex. This complex is electrochemically detectable and distinguishable from the Ru(II) precursor. At short times, the reaction follows first-order kinetics with a rate constant 10 times greater than for the equivalent homogeneous reaction. The reaction rate is proportional to PN_2 up to 8 atm and possibly higher. The sensitivity approaches 1/10 atmosphere and the minimum electrode response time at elevated PN_2 is 10 sec. This N_2 sensing system holds promise for eventual configuration into an implantable unit for in situ measurements of tissue PN_2 during compression and decompression of an experimental animal.

LIST OF FIGURES

	<u>Page</u>
Fig. 1 Reaction of $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ (1 mM in 0.1M H_2SO_4) with N_2 (1 atm)	6
Fig. 2 Reaction of $\text{cis-}[\text{Ru}(\text{en})_2(\text{H}_2\text{O})_2]^{+2}$ (1 mM in 0.1M H_2SO_4) with N_2 (1 atm)	7
Fig. 3 Reaction of $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ (5 mM) with pyri- dine (12.36 mM) at pH 6.8 (Ar-saturated sulfate buffer	10
Fig. 4 Reaction of $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ (5 mM) with pyridine (12.36 mM) at pH 1.5 (Ar-saturated 0.1M H_2SO_4)	11
Fig. 5 Reaction of $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ (5 mM) with pyri- dine (12.36 mM) and N_2 1 atm, pH = 1.5	12
Fig. 6 Formation of $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})\text{py}]^{+2}$ at pH 1.5 in presence of Ar and N_2	13
Fig. 7 Rate of disappearance of $\text{Ru}(\text{en})_2(\text{H}_2\text{O})_2^{2+}$ at -0.17V vs. SCE and $\text{Ru}(\text{en})_2(\text{py})(\text{H}_2\text{O})_2^{2+}$ at 0.05V vs. SCE in N_2 saturated 0.1M H_2SO_4	15
Fig. 8 Variation in reaction rate for $\text{Ru}(\text{II})\text{-N}_2$ formation during equilibrium of solution PN_2	22

1.0 INTRODUCTION

This report describes work carried out under Phase II of Office of Naval Research-Defense Small Business Advanced Technology Contract N00014-83-C-0458, during the period June 15, 1983 to June 14, 1986. The three year program of research was undertaken to develop an electrochemical sensor for quantitating PN_2 in biological tissues and fluids. Because molecular dinitrogen is not readily oxidized or reduced in an aqueous solvent, the key to developing an electrochemical sensor for dinitrogen was to find a reaction involving dinitrogen whose product could be detected electrochemically. The findings that certain transition metal ions react in aqueous solution with molecular dinitrogen (1-3) provided the impetus for this work. Results obtained in the Phase I portion of the program under Contract No. N00014-82-C-0133 demonstrated the feasibility of using transition metal-dinitrogen chemistry as a basis for a N_2 sensor (4). That program identified Ru pentaammine dinitrogen as a potential system for a N_2 sensor, and illustrated how a reaction sequence cycling from Ru(II) to Ru(II)-dinitrogen to Ru(III)-dinitrogen (unstable) and back to Ru(II) might be used for measuring PN_2 .

The specific objectives of the Phase II research program were to identify other transition metal ion-dinitrogen complex systems whose chemical and electrochemical properties would be favorable for use in quantitating PN_2 , and to develop methods for immobilizing those metal ions so that the nitrogen complexing and electrochemical reactions of interest would take place on an electrode surface under physiological conditions.

To accomplish these objectives, the research program was organized into four tasks:

1. Perform basic studies of selected transition metal dinitrogen complexes including the kinetics of their formation from molecular dinitrogen and their electrochemical properties.
2. Prepare derivatized electrodes containing the dinitrogen complexing transition metal ions whose properties appeared most favorable for developing a N_2 sensing electrode.
3. Evaluate derivatized N_2 sensing electrodes for their ability to measure PN_2 under hyperbaric conditions and in simulated physiological solutions.

4. Develop appropriate electroanalytical methodology to obtain rapid and sensitive iterative measurements of P_{N_2} .

The three year program culminated in an electrochemical sensor for dissolved nitrogen which utilizes the reaction of Ru(II) ammine or ethylenediamine cations with molecular N_2 in solution to measure the concentration of dissolved N_2 . As presently configured, the N_2 sensor is constructed of a glassy carbon electrode which is coated with an ion exchange polymer membrane (Nafion), containing one of the following Ru(III) cations: Ru pentaammine, cis-Ru tetraammine or cis-Ru ethylenediamine. The reduced form of the Ru cation on the electrode surface reacts with N_2 in solution to form a Ru(II)- N_2 complex. This complex is electrochemically detectable and distinguishable from the Ru(II) precursor. The concentration of N_2 in solution, P_{N_2} , is quantified by measuring the rate of the reaction between the surface attached Ru(II) cation and the N_2 in solution. The details of the preparation and evaluation of this N_2 sensor are provided in a manuscript appended to this report, "Development of a N_2 Sensor for Determination of P_{N_2} in Body Tissue," by T. J. Lewis and L. S. Robblee (5). The main body of the report which follows focuses on the solution studies of dinitrogen complex formation, the electrochemical properties of dinitrogen complexes, and the alternative immobilization strategies which were evaluated during the development of the N_2 sensor.

2.0 SOLUTION STUDIES OF DINITROGEN COMPLEX FORMATION

2.1 Survey of Transition Metal Dinitrogen Complexing Cations

2.1.1 Ruthenium

A number of Ru(III) compounds were surveyed to identify those which react with N_2 in aqueous solution to yield an electrochemically detectable product. The Ru(III) compounds were reduced with Zn amalgam under Ar saturation and then transferred to N_2 saturated electrolyte for in situ formation of Ru(II)- N_2 complexes. Cyclic voltammetry was used to detect the formation of N_2 complexes and to determine the oxidation potentials of those complexes.

Table 1 lists the Ru compounds tested, their oxidation and reduction potentials, and the potentials at which current peaks associated with dinitrogen complexes were observed. Electrochemical potentials are referenced to a saturated calomel electrode (SCE). Of the compounds tested, cyclic voltammetry of the cis-dichloro, dibromo, and diaquo tetraammines indicated that these compounds reacted with N_2 in a similar manner to produce three new oxidative current peaks in the cyclic voltammograms. Dinitrogen complex formation was also observed with cis-Ru ethylenediamine, although cyclic voltammetry demonstrated only one or two new oxidative current peaks, rather than three. No dinitrogen complex formation was observed with the trans-dichloro tetraamine, cis-sulfatochlorotetraamine or cis-dichloro-bis-pyridine tetraamine complexes of Ru.

Based on these results, the cis-Ru tetraamine and cis-Ru ethylenediamine complexes were selected for further study as candidate materials for a N_2 sensor, in addition to the Ru pentaamine complex which was identified as a candidate material during Phase I of this program (4). Characteristic of the Ru dinitrogen electrochemistry is the absence of a reduction peak in the cyclic voltammogram for reduction of Ru(III)- N_2 back to Ru(II)- N_2 because of the rapid loss of N_2 from the Ru(III)- N_2 species. The literature value for the rate of loss of N_2 from $[Ru(NH_3)_5N_2]^{+2}$ is $\gg 50 \text{ sec}^{-1}$ (6). The instability of Ru(III)- N_2 is favorable for utilization of Ru dinitrogen complex chemistry in a N_2 sensor since the precursor N_2 complexing cation can be regenerated electrochemically after oxidation of the N_2 complex.

2.1.2 Osmium

Two osmium compounds, $[Os(NH_3)_5N_2]Cl_2$ and $[Os(NH_3)_5Cl]Cl_2$ were synthesized according to literature methods (2,7) and their electrochemical properties evaluated by cyclic voltammetry. Voltammetry of the

TABLE 1
CYCLIC VOLTAMMETRIC DATA FOR RUTHENIUM COMPOUNDS
AND RUTHENIUM-DINITROGEN COMPLEXES

Compound	Electrolyte	Redox Potentials (mV) of $\text{Ru}^{+3} \rightleftharpoons \text{Ru}^{+2}$		E_{ox} of N_2 Complex(es) (mV)
		E_{OX}	E_{RED}	
cis-[Ru(NH ₃) ₄ Cl ₂]Cl	Acid ¹	-140	-340	
	Acid	-120	-280	+500
	+ Zn Amalgam			+700 +900
cis-[Ru(NH ₃) ₄ Cl ₂]Cl	Buffer ²	-130	-305 (major)	+450
	+ Zn Amalgam		-210 (shoulder)	+600 +725
cis-[Ru(NH ₃) ₄ Br ₂]Br	Buffer	-145	-380	
	Buffer	-140	-320 (major)	+500
	+ Zn Amalgam		-280 (shoulder)	+700 +825
cis-[Ru(NH ₃) ₄ (H ₂ O) ₂ (TFMS) ₃	Buffer	-195	-375 (major)	
			-280 (shoulder, 1st scan only)	
	Buffer	-180	-370	+500
cis-[Ru(NH ₃) ₄ ClSO ₂]Cl	Buffer	+280 (major)	+170 (major)	
		-200 (minor)	-330 (minor)	
	Buffer	+130 (major)	+ 50 (major)	Ambiguous
	+ Zn Amalgam	-200 (minor)	-340 (minor)	
trans-[Ru(NH ₃) ₄ Cl ₂]Cl		+680 (incr. in Ar; decreased in N ₂)	+550 (incr. in Ar; decreased in N ₂)	
	Buffer	1st cycle: -300 to -200	-450	
		2nd cycle: -300 (shoulder) -380 (peak)	-450 -250 (shoulder)	
	Buffer	-200 (major)	-350 (major)	None
		-550 (appears on multiple cycles & shifted to -400 mV under N ₂)	-650 (appears in multiple cycles) and seemed to increase under N ₂	
cis-[Ru(en) ₂ Cl ₂]Cl	Buffer	-125	-350	
	Buffer + Zn Amalgam	- 50	-400	+475 +780
cis-[Ru(en) ₂ Br ₂]Br	Acid	-120	-375	
	Acid + Zn Amalgam	- 80	-280	plateau 600-850
cis-[Ru(bipy) ₂ Cl ₂]Cl	Buffer	+455	+320	
	Buffer + Zn Amalgam	+575	+380	None

¹Acid = 0.1M H₂SO₄

²Buffer = 0.001M H₂SO₄-0.099M K₂SO₄, pH 3.3

$[\text{Os}(\text{NH}_3)_5\text{Cl}]^{+2}$ cation demonstrated a reduction potential at -1.15V vs. SCE which is too cathodic to be useful in a physiological environment. Voltammetry of the dinitrogen complex demonstrated an oxidation wave at 0.350V vs. SCE with a corresponding reduction wave at 0.180V vs. SCE. The reversibility of the $[\text{Os}(\text{NH}_3)_5\text{N}_2]^{+2/+3}$ couple was consistent with a higher degree of stability of the Os(III)- N_2 complex than was observed for any of the Ru dinitrogen complexes studied. The rate constant for loss of N_2 from Os(III)pentaamminedinitrogen is $2 \times 10^{-2} \text{sec}^{-1}$ (8). In view of these properties of the osmium amines, e.g., the extreme cathodic potential for the Os(II/III) couple and the slow dissociation of N_2 from the Os(III)- N_2 complex, we concluded that a N_2 sensor could not be based on osmium - dinitrogen chemistry, and concentrated the research effort on the chemistry and electrochemistry of the Ru ammine and ethylenediamine complex.

2.2 Kinetics of Dinitrogen Complex Formation

Kinetic studies of the formation of dinitrogen complexes of Ru tetraammine and Ru(bis)ethylenediamine were carried out using cyclic voltammetry as described previously for Ru pentaammine dinitrogen (4). The Ru(III) precursors in argon-saturated electrolyte were reduced with Zn/Hg in a separatory funnel having a stopcock connection to the electrochemical cell. When the reduction was complete (75-90 minutes as determined in preliminary cyclic voltammetry experiments), the reduced Ru(II) precursor was admitted to N_2 -saturated electrolyte in the electrochemical cell.

The progress of the reaction between the Ru(II) precursors and N_2 are illustrated in the cyclic voltammograms of Figures 1 and 2. The first cycle in each set of voltammograms shows a single anodic peak at -0.12V vs. SCE representing the oxidation of the precursor compounds, $\text{cis}-[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ (Figure 1) and $\text{cis}-[\text{Ru}(\text{en})_2(\text{H}_2\text{O})_2]^{+2}$ (Figure 2). As the reactions proceeded, the magnitude of these two current peaks decreased due to the complexation of the precursor compounds with N_2 . At the same time, 2 or 3 new anodic peaks appeared at higher potentials representing the oxidation of the newly-formed dinitrogen complexes.

The same sequence of reactions was observed with the dichloro, dibromo and diaquo salts of Ru(III) tetraammine as starting materials, in both pH 3.3 sulfate buffer and in acid electrolyte. In the acid electrolyte, the anodic current peaks associated with dinitrogen complex oxidation were more sharply defined and occurred at slightly higher potentials than they were in the pH 3.3 buffer.

The dibromo and dichloro Ru ethylenediamine compounds differed from the tetraamines and from each other in the location of anodic current peaks associated with N_2 complex formation. With the dichloro form as the starting material, two anodic peaks at 0.475V and 0.78V vs. SCE were observed to increase in magnitude as the reaction with N_2

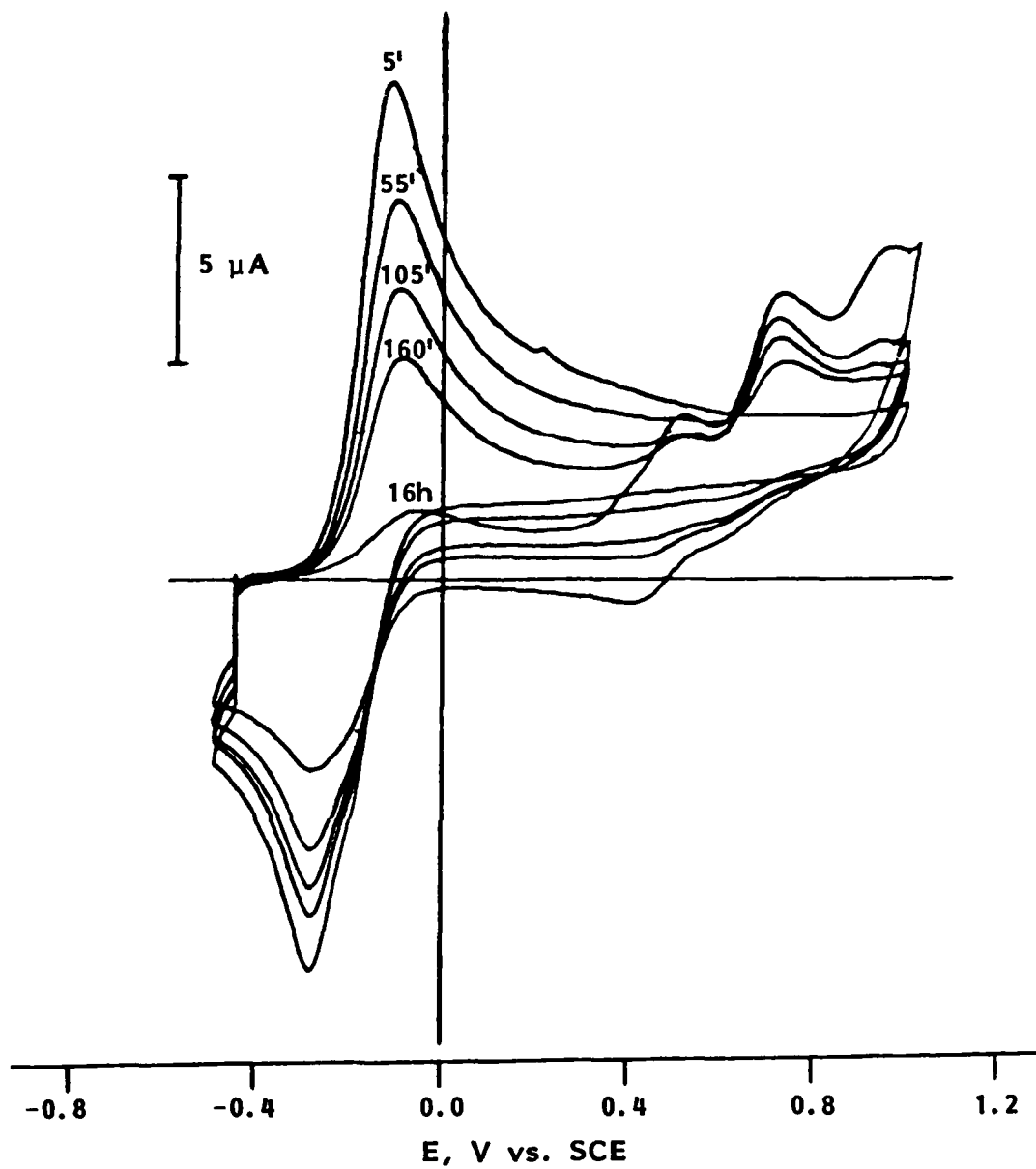


Fig. 1. Reaction of $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{2+}$ (1 mM in 0.1M H_2SO_4) with N_2 (1 atm). Cyclic voltammograms were recorded at different times during the reaction.

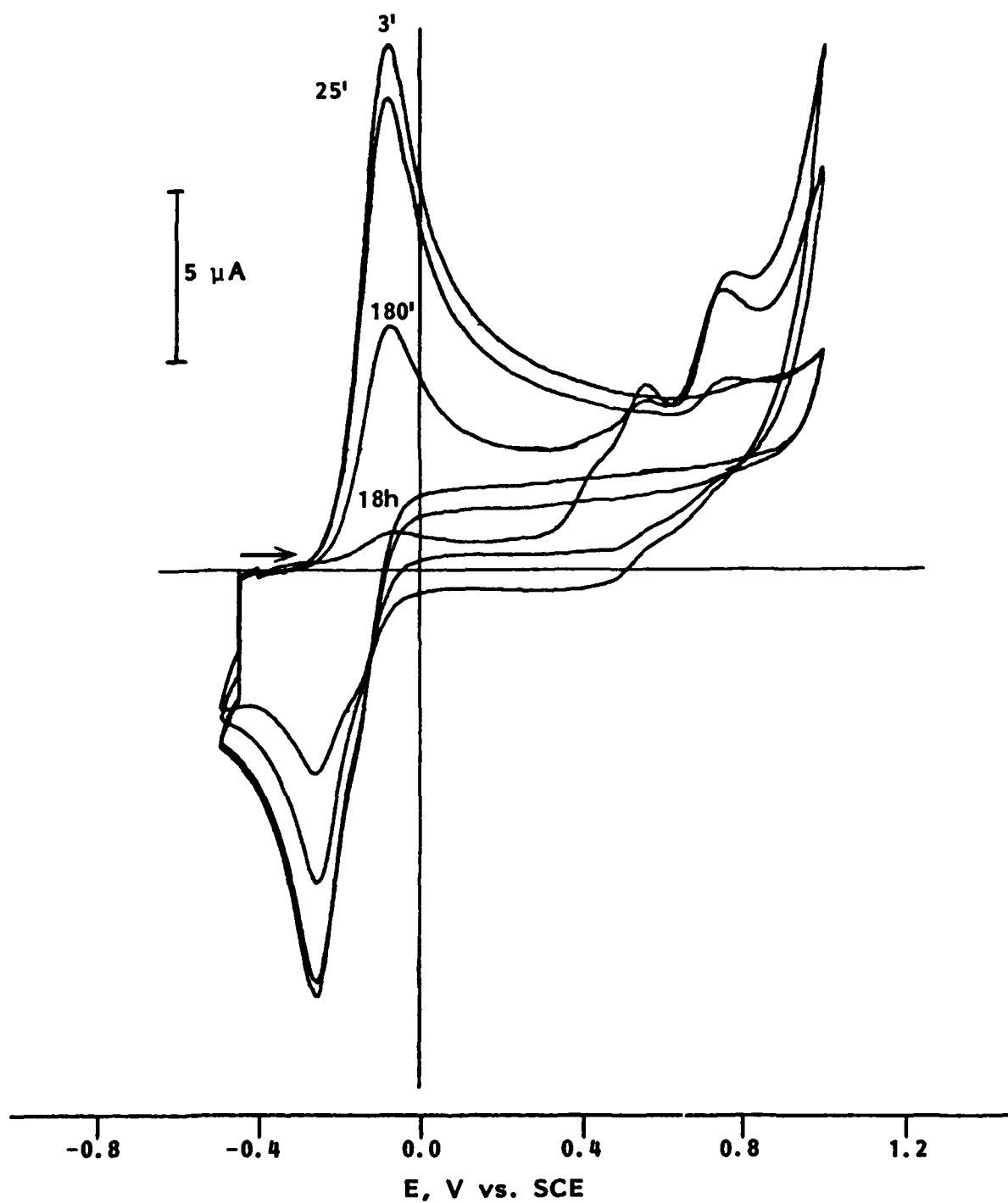
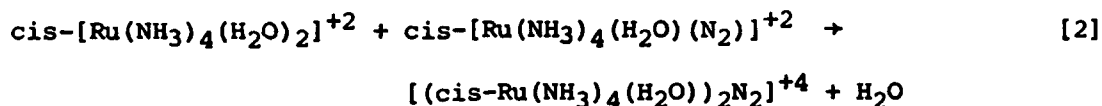
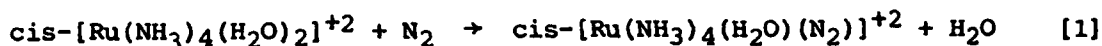


Fig. 2. Reaction of $\text{cis-}[\text{Ru}(\text{en})_2(\text{H}_2\text{O})_2]^{2+}$ (1 mM in 0.1M H_2SO_4) with N_2 (1 atm). Cyclic voltammograms were recorded at different times during the reaction.

progressed. A cathodic peak at $\sim 0.4V$ vs. SCE also increased in magnitude. With the dibromo Ru ethylenediamine, a single broad plateau of anodic current between 0.6-0.85V vs. SCE was observed after prolonged exposure to N_2 . It is not known at the present time why these two ethylenediamine compounds behave differently toward dinitrogen.

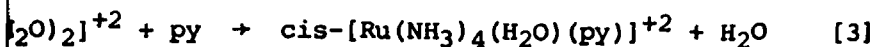
The current (i_p) in the anodic peaks at $-0.12V$ vs. SCE provided a direct measure of the Ru(II) precursor concentrations at any time during the reaction with N_2 . For two reactions carried out with 1 mM concentration of each Ru(II) precursor, plots of $\ln(i_p)$ vs. time were linear for 75-100 minutes with pseudo-first order rate constants (k') of $7.61 \pm 1.22 \times 10^{-5} \text{ sec}^{-1}$ and $8.78 \pm 0.11 \times 10^{-5} \text{ sec}^{-1}$ for the formation of $[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})(\text{N}_2)]^{+2}$ and $[\text{Ru}(\text{en})_2(\text{H}_2\text{O})(\text{N}_2)]^{+2}$, respectively. These are about twice what was found for the formation of $[\text{Ru}(\text{NH}_3)_5\text{N}_2]^{+2}$. With 5 mM concentration of $[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$, plots of $\ln(i_p)$ vs. time showed two linear segments, the first extending up to 30 minutes with $k' = 6.8 \pm 0.98 \times 10^{-5} \text{ sec}^{-1}$, and the second extending up to 150 minutes with an increased slope of $1.038 \pm 0.01 \times 10^{-4} \text{ sec}^{-1}$. The change in slope of the $\ln(i_p)$ vs. t plots coincided with the appearance in the cyclic voltammogram of an anodic peak at 0.5V vs. SCE, which was probably due to the oxidation of a binuclear species $[(\text{cis-Ru}(\text{NH}_3)_4(\text{H}_2\text{O}))_2\text{N}_2]^{+4}$. The increased slope after 30 minutes of reaction was consistent with the participation of $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ in two reactions:



2.3 Reaction of cis-Ru Tetraammine with Pyridine

One mechanism considered for immobilizing Ru complexes to an electrode surface was ligand substitution of pyridine (py) for one of the aquo ligands of $\text{cis-Ru(II) diaquotetraammine}$, $\text{cis-Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ or $\text{cis-Ru(II) diaquo-bis-ethylenediamine}$, $\text{cis-}[\text{Ru}(\text{en})_2(\text{H}_2\text{O})_2]^{+2}$ (9). Accordingly, several experiments were performed to determine the optimum conditions for forming the monosubstituted py compound and to obtain electrochemical data for subsequent use in evaluating the immobilization procedures.

The reaction between $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ and py was carried out in the same manner as the dinitrogen reactions, with cyclic voltammetry being used to monitor the formation of py complexes. As reported in the literature, the reaction proceeds at pH 7 via two successive substitution reactions (9):



formation of both products was readily observed by cyclic voltammetry at the different oxidation potentials of reactants and products. In Figures 3 and 4, pH had a marked effect on the reaction. At pH 6.8 (Figure 3), both monosubstituted (IIa & IIc) py complexes were predominant as the reaction proceeded. At pH 4 (Figure 4), only the monosubstituted product (IIa and IIc) was observed after 315 minutes of reaction. This was believed to be a consequence of the eventual immobilization of Ru tetraammine to the electrode surface via py groups where attachment of Ru through only one py group would be the preferred mechanism.

(II) tetraammine reaction with py was carried out. In cyclic voltammetry demonstrated the formation of the monosubstituted py complex as well as three dinitrogen complexes. The magnitude of the cathodic current peak at 0.0V vs. SCE decreased with the reduction of the monosubstituted py complex, cis- $[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})(\text{py})]^{+2}$ led to follow its formation during the reaction. The current at 0.0V vs. SCE with time is shown in Figure 6 for Ru-py and N_2 saturated solutions. The two curves were the same initially. After that time, the current attributed to the reduction of the monosubstituted py complex led to increase in Ar-saturated solution. However, in N_2 solution, the current reached a maximum level in 120 minutes and then leveled off slightly at longer reaction times. This leveling off of the current for cis- $[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})(\text{py})]^{+2}$ reduction suggested that the monosubstituted py complex ceased, possibly because of diminished free Ru tetraammine. An alternative possibility was the reduction of the py complex followed by its reaction with N_2 to form cis- $[\text{Ru}(\text{NH}_3)_4(\text{py})\text{N}_2]^{+2}$. Several observations tended to support the latter possibility.

The plot of $\ln i_{pa}$ (-0.12V vs. SCE) vs. t for the decrease in the current for cis- $[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ were linear only for 30-40 minutes with a pseudo first order rate constant, k' , of $2.2 \times 10^{-4} \text{ sec}^{-1}$. This was higher than the rate constant for the reduction of the monosubstituted py complex with py alone ($1.01 \times 10^{-4} \text{ sec}^{-1}$) consistent with the formation of the Ru complex in two reactions. However, as the reaction proceeded, the $\ln i_{pa}$ vs. t plot became curvilinear with a decreasing slope. The slower rate of cis- $[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ reduction with a continuing increase in anodic currents suggested that these anodic peaks represent the reduction of mixed py- N_2 complexes as well as complexes containing both py and N_2 . It is likely that cis- $[\text{Ru}(\text{NH}_3)_4(\text{py})\text{N}_2]^{+2}$ and cis- $[\text{Ru}(\text{NH}_3)_4(\text{py})_2]^{+2}$ have similar oxidation potentials so that they could be reduced solely by electrochemical means.

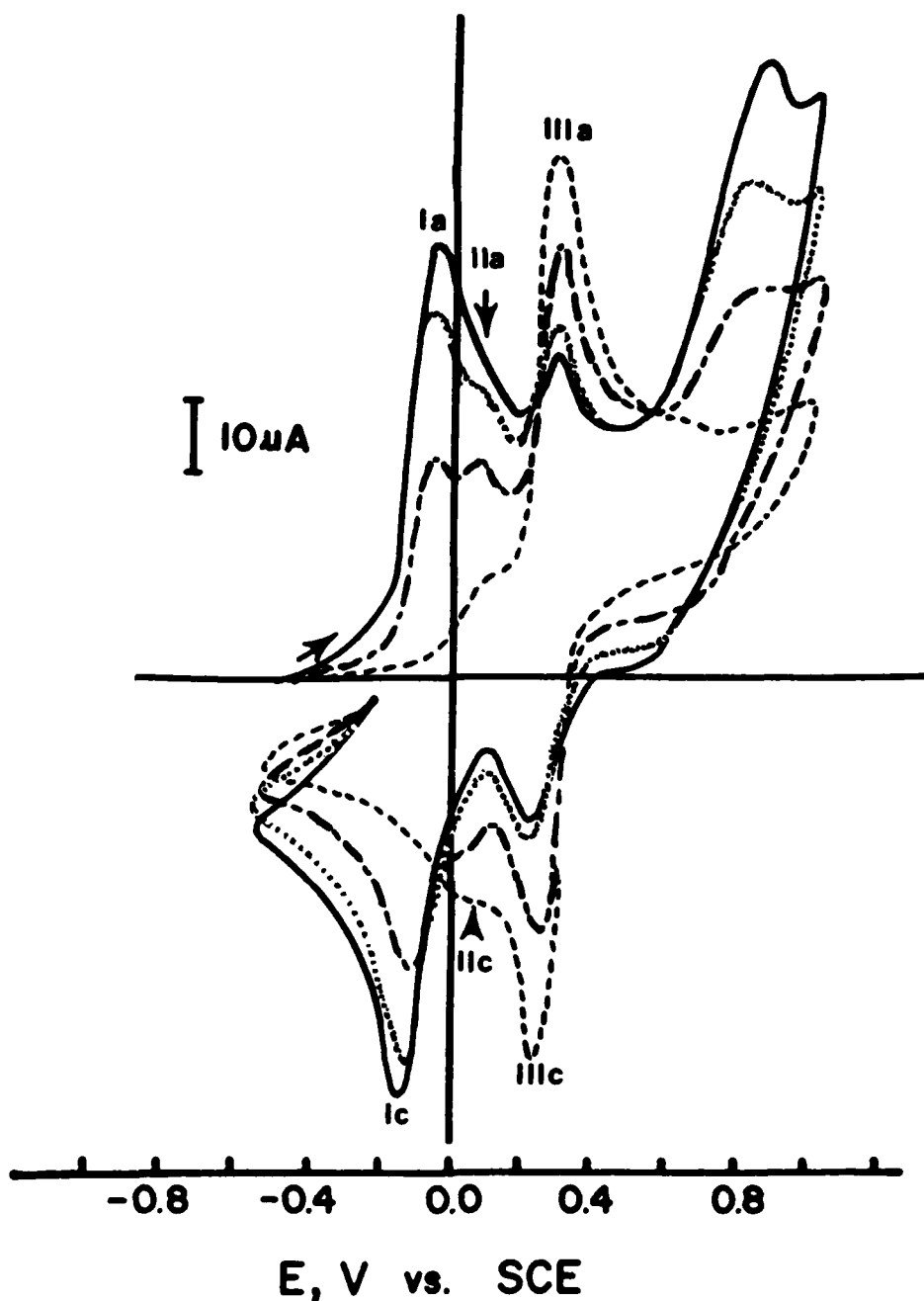


Fig. 3. Reaction of $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ (5 mM) with pyridine (12.36 mM) at pH 6.8 (Ar-saturated sulfate buffer). Cyclic voltammograms were recorded at different times during the reaction: 5 min (—); 20 min (...); 82 min (-·-); 16 hr (---).

Peaks Ia and Ic: $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2} \rightleftharpoons +3$
 Peaks IIa and IIc: $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})(\text{py})]^{+2} \rightleftharpoons +3$
 Peaks IIIa and IIIc: $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{py})_2]^{+2} \rightleftharpoons +3$

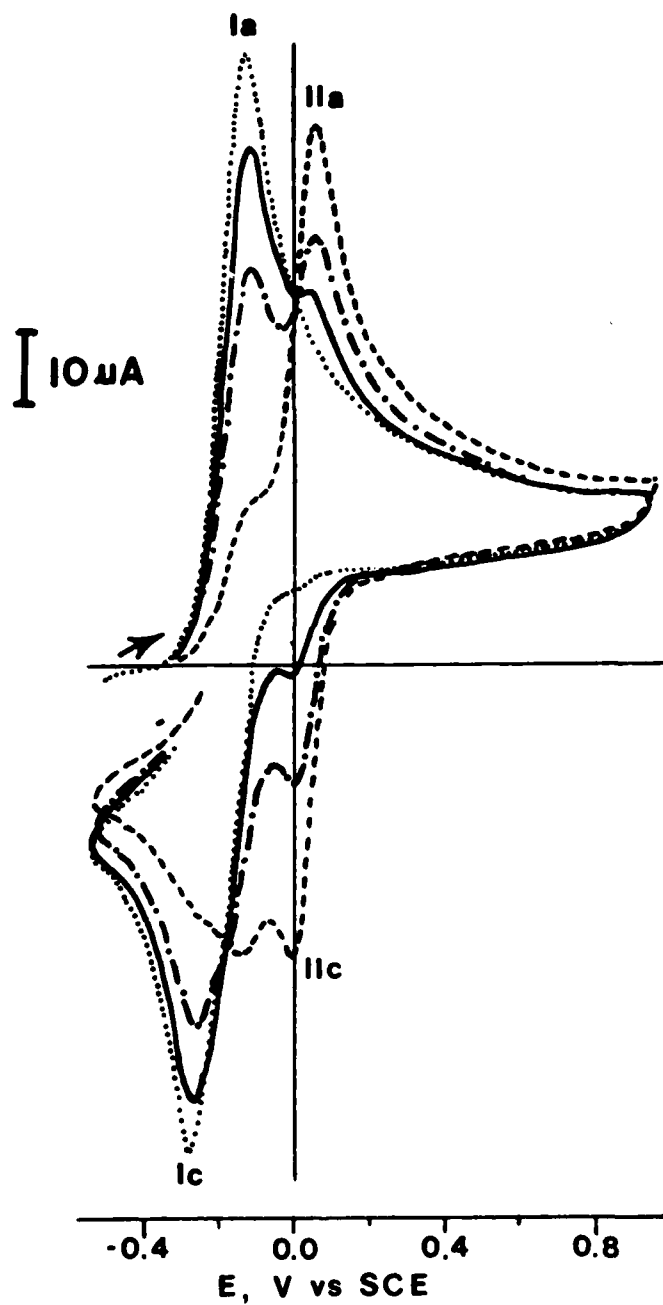


Fig. 4. Reaction of $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ (5 mM) with pyridine (12.36 mM) at pH 1.5 (Ar-saturated 0.1M H_2SO_4). Cyclic voltammograms were recorded at different times during the reaction: 2 min (\cdots); 27 min (—); 80 min (---); 315 min (- - -).
 Peaks Ia and Ic: $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2} \rightleftharpoons +3$
 Peaks IIa and IIc: $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})(\text{py})]^{+2} \rightleftharpoons +3$

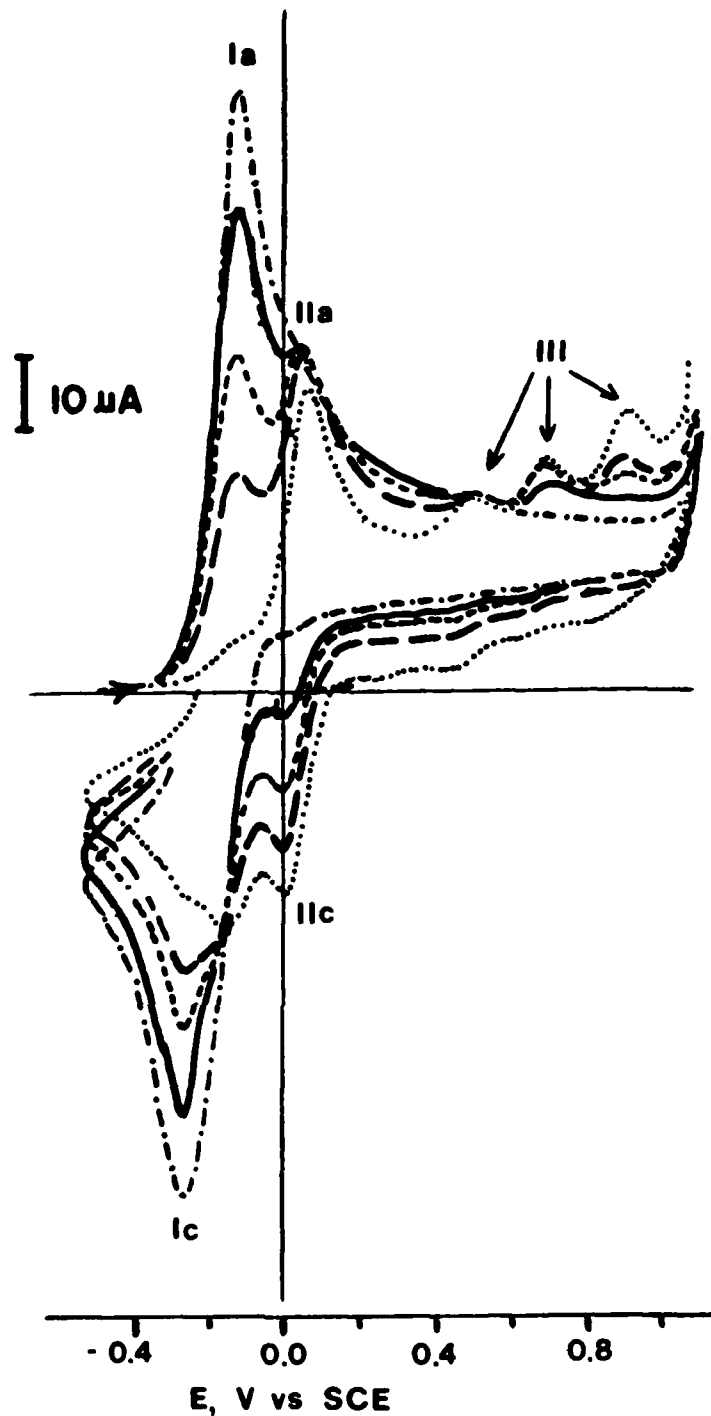


Fig. 5. Reaction of $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2}$ (5 mM) with pyridine (12.36 mM) and N_2 1 atm, pH = 1.5. Cyclic voltammograms were recorded at different times during the reaction: 0' (-.-.); 18' (—); 50 min (---); 110 min(- - -); 320 min (···).

Peaks Ia and Ic: $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{+2} \rightleftharpoons +3$
 Peaks IIa and IIc: $\text{cis-}[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})(\text{py})]^{+2} \rightleftharpoons +3$
 Peaks III: Oxidation of dinitrogen complexes

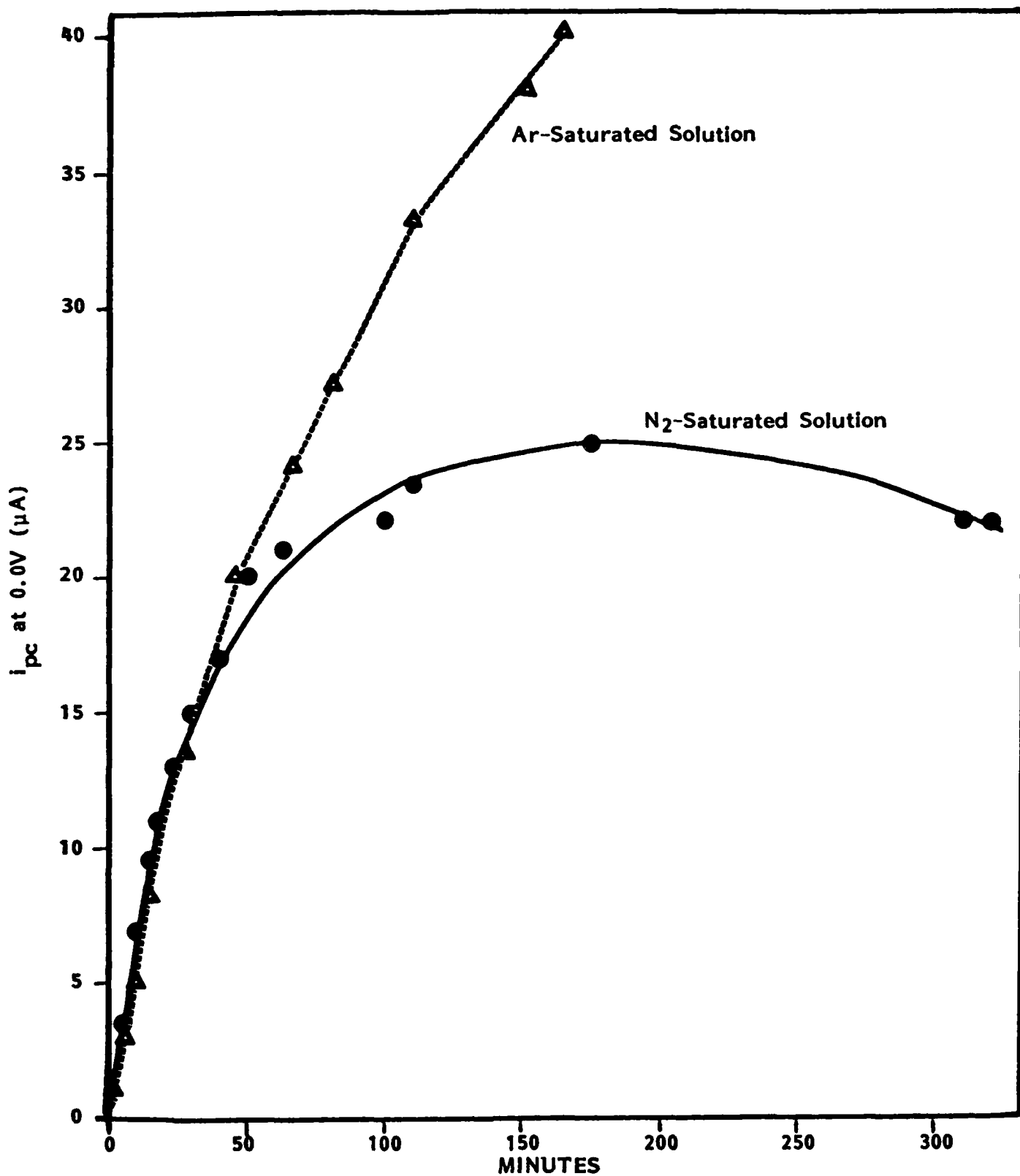


Fig. 6. Formation of $cis-[Ru(NH_3)_4(H_2O)py]^{+2}$ at pH 1.5 in presence of Ar and N_2 . Value for i_{pc} determined by cyclic voltammetry as illustrated in Figures 3 and 4.

2.3.1 Reaction of cis-Ru Pyridyltetraammine with N₂

Although the results discussed in Section 2.3 suggest the formation of a disubstituted pyridyl-dinitrogen complex of cis-Ru tetraammine, nevertheless, subsequent results demonstrated a very limited substitution of N₂ into Ru complexes bound to surfaces by immobilized py groups (Section 3.2). To reconcile these differences, we embarked on a series of experiments exploring the rate of N₂ binding by cis-[Ru(en)₂(H₂O)py]⁺² in aqueous solution and comparing it to the rate of N₂ binding by the diaquo complex, cis-[Ru(en)₂(H₂O)₂]⁺².

To evaluate the relative rates of N₂ complex formation, a 1 mM solution of cis-[Ru(en)₂(H₂O)₂]⁺² was prepared in 0.1M H₂SO₄ by Zn/Hg reduction of cis-[Ru(en)₂(Cl)₂]⁺¹ under an Ar purge. After separating the reductant, 1 mole equivalent of py was added to the solution of the Ru(II) cation and the reaction allowed to run to completion. The formation of the monosubstituted py complex was monitored by cyclic voltammetry. Overnight stirring under Ar was required for complete conversion of the diaquo species to the py complex. Next, N₂ gas was bubbled through the solution and the formation of the N₂ complex was followed by the decrease in the current peak at 0.05V vs. SCE for the oxidation of the Ru(II)-py complex, and an increase in a current peak at 0.8-0.9V vs. SCE for the oxidation of a Ru(II)-py-N₂ complex. After three days of reaction, there was only a 25% conversion of the Ru(II)-py complex to Ru(II)-py-N₂. In contrast, a parallel reaction between cis-[Ru(en)₂(H₂O)₂]⁺² and N₂ achieved equilibrium within 5-6 hours (Figure 7).

The slow reaction between N₂ and cis-[Ru(en)₂(H₂O)py]⁺² compared to cis-[Ru(en)₂(H₂O)₂]⁺² was confirmed through a competitive reaction experiment in which both reactants were present in equimolar concentrations. The results of these studies, extrapolated to the surface-confined cis-[Ru(en)₂(H₂O)py]⁺², suggested that the strong metal-to-ligand π interaction between py and Ru(II) interferes with the π interaction with N₂ that is necessary for coordination of N₂ to the metal center.

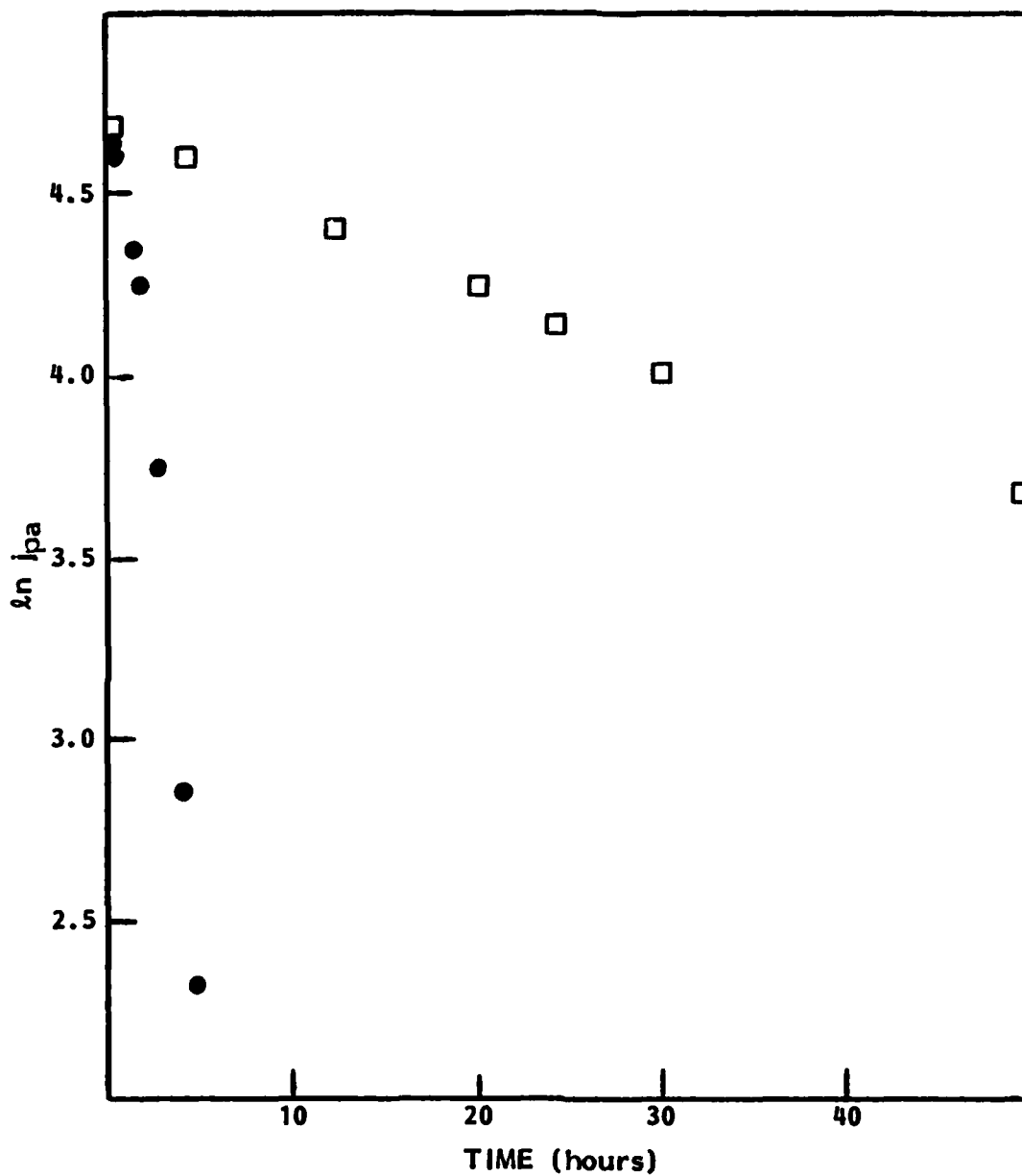


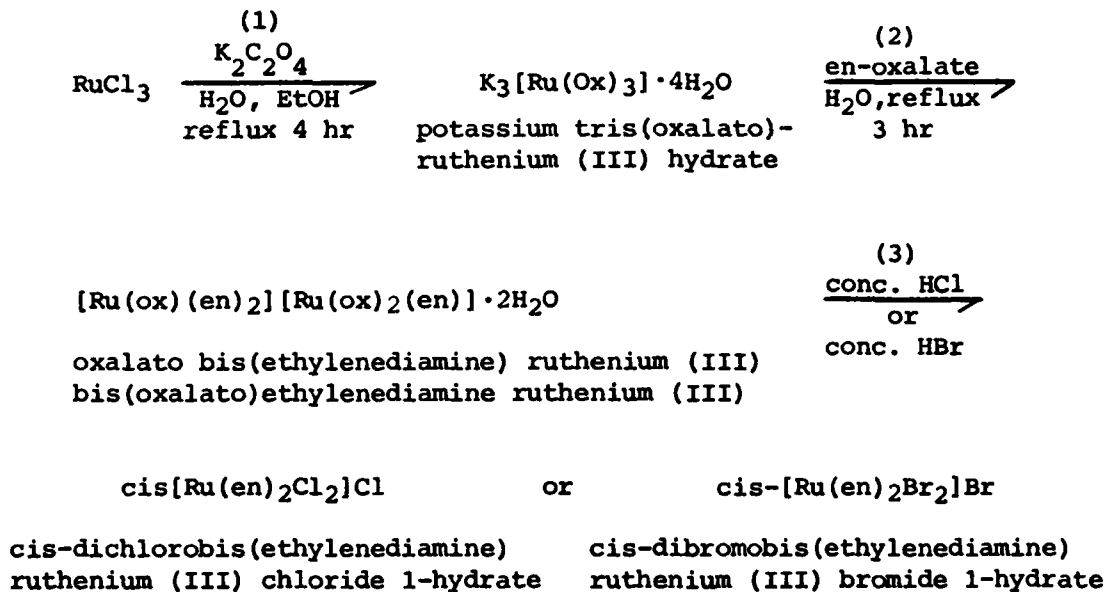
Fig. 7. Rate of disappearance of $\text{Ru}(\text{en})_2(\text{H}_2\text{O})_2^{2+}$ (●) at -0.17V vs. SCE and $\text{Ru}(\text{en})_2(\text{py})(\text{H}_2\text{O})_2^{2+}$ (□) at 0.05V vs. SCE in N_2 saturated 0.1M H_2SO_4 . j_{pa} = current density for the respective anodic peak.

3.0 IMMOBILIZATION OF Ru CATIONS TO ELECTRODE SURFACES

Several different procedures were investigated to attach a N₂-complexing Ru(II) cation to an electrode surface. These fell into four general categories: 1) in situ synthesis of the Ru bis-ethylenediamine complex on an ethylenediamine-silane derivatized electrode surface; 2) attachment of Ru complexes by a ligand substitution reaction with surface pyridine groups; 3) functionalization of the Ru complex with py-silane or poly-4-vinylpyridine (PVP) prior to attachment to the electrode; and 4) noncovalent binding by electrostatic attraction to an ion exchange polymer.

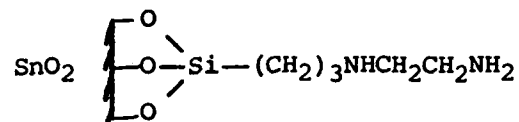
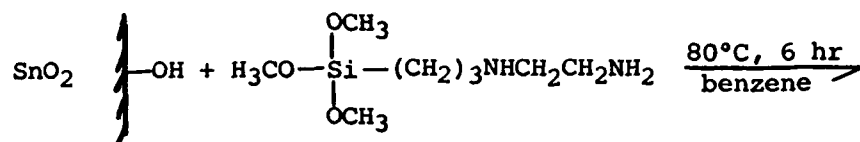
3.1 In Situ Synthesis of cis-Ru bis-ethylenediamine on SnO₂-Coated Electrodes

The preparation of the bis-ethylenediamine Ru(III) complex in solution is a three-step synthesis (10):

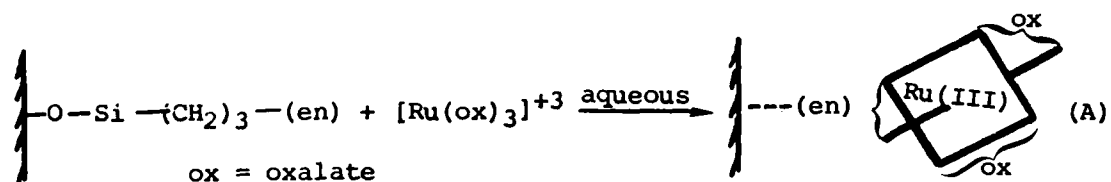


The reaction scheme for carrying out the synthesis of an electrode surface was adapted from the solution reaction as follows:

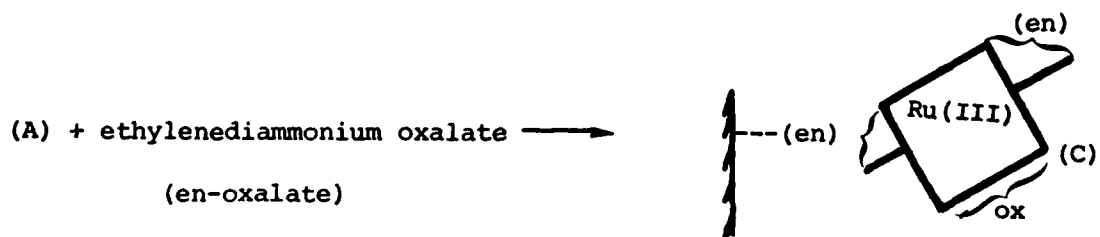
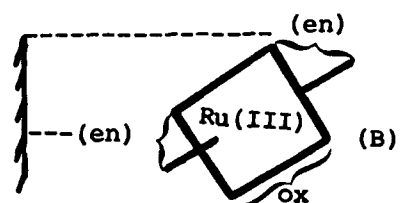
1. Derivatize an SnO₂ electrode with ethylenediamine ligands employing organosilane chemistry (11):



2. React the en-silane-SnO₂ electrode in a solution of K₃[Ru(ox)₃]·4H₂O to substitute the ethylenediamine ligand(s) for the oxalato ligands resulting in attachment of Ru centers via coordination to ethylenediamine:



and/or



3.3 Functionalization of Ru Complexes

The chloride salts of Ru tetraammine and bis-ethylenediamine were dissolved in dry methanol, reduced with Zn/Hg, and reacted in methanol with dissolved py-silane or PVP. The resulting py-substituted complexes were applied to an electrode surface by transfer in methanol, followed by solvent evaporation. The silane-containing complex was "annealed" to the electrode by contact with H₂O which promotes silane hydrolysis. This "methanol" synthesis had the advantages of 1) requiring less Ru salts for a given number of electrodes, and 2) requiring less time than the soaking method described in Section 3.2. However, these py-derivatized electrodes suffered the same problems of limited N₂ binding and very slow rates of N₂ binding as did the electrodes prepared by soaking py-derivatized electrodes in Ru solutions.

3.4 Electrostatic Binding of Ru Cations

Since the covalent binding of Ru dinitrogen complex precursors to an electrode surface was unsatisfactory for our purposes, a noncovalent method was investigated as it would cause less perturbation of the electronic structure of the Ru center. Of the noncovalent binding mechanisms possible, adsorption and electrostatic attraction, the latter seemed most suitable for the Ru ammine and ethylenediamine complexes which have multiple cationic charge per molecule.

The ion exchange polymer, Nafion (Dupont), is a perfluorosulfonate polymer composed of a polytetrafluoroethylene (PTFE) backbone and pendant side chains terminated with -SO₃ M⁺. The polymer is available from commercial sources as a dry film or as a lower molecular weight polymer dissolved in alcohol. The polymer was applied to a glassy carbon (GC) electrode surface by micropipette transfer of 1 μl of a 0.5% (wt/vol) alcohol solution (thin film) or 0.5 μl of a 5% (wt/vol) solution (thick film) followed by solvent evaporation. Once dried, the polymer is essentially irreversibly bound to the electrode surface.

A Ru(III) cation was incorporated into the Nafion film by soaking the GC/Naf electrode in a dilute aqueous solution containing the appropriate Ru(III) cation, generally 10-25 μM Ru in 0.001-0.1M supporting electrolyte. Ru(III) cation uptake into the polymer was monitored by cyclic voltammetry. Equilibrium was established between the solution and the Nafion film within about 10 minutes for thick films, and even shorter times for thin films.

3.4.1 Electrochemistry of Nafion-Bound Ru

The cyclic voltammetric response of Nafion-bound Ru cations in blank electrolyte was consistent with reversible, surface-confined reactants for thin films of Nafion (13). Anodic and cathodic peaks were only slightly separated (E_p = 0-20 mV) and the peak width at half

height was 90 mV. The peak potentials for the Ru +3 \rightleftharpoons +2 conversions were centered about -0.3V vs. SCE, slightly more negative than observed in solution studies due to strong electrostatic interaction with the Nafion polymer (14).

3.4.2 Stability of Nafion-Modified Electrodes

Shelf life. Both the plain GC/Naf electrodes and the GC/Naf electrodes with bound Ru(III) cation (GC/Naf-Ru(III) electrodes) can conveniently be prepared in advance and stored several days or longer in a clean, dry atmosphere without any deterioration of properties. Preparation for use requires only a short period of hydration in distilled water or dilute electrolyte.

Stability of GC/Naf-Ru electrodes in solution. During exposure to electrolyte solution, some loss of Ru occurs through ion exchange or partition with the solution. Efflux was monitored with cyclic voltammetry using the magnitude of the current peaks at -0.30V vs. SCE as a direct measure of Ru concentration. Factors affecting the efflux rate were the following:

a) Ru cation. No differences were observed between the tetraammine, pentaammine or ethylenediamine complexes of Ru in terms of efflux rate. However, the +3 oxidation state of the Ru center of all three complexes was much more stably bound than the reduced +2 cation. This is fortuitous for the actual use of the Nafion-bound Ru cation as a N₂ sensor since the Ru center is maintained in the +3 state except for short times when making a PN₂ measurement.

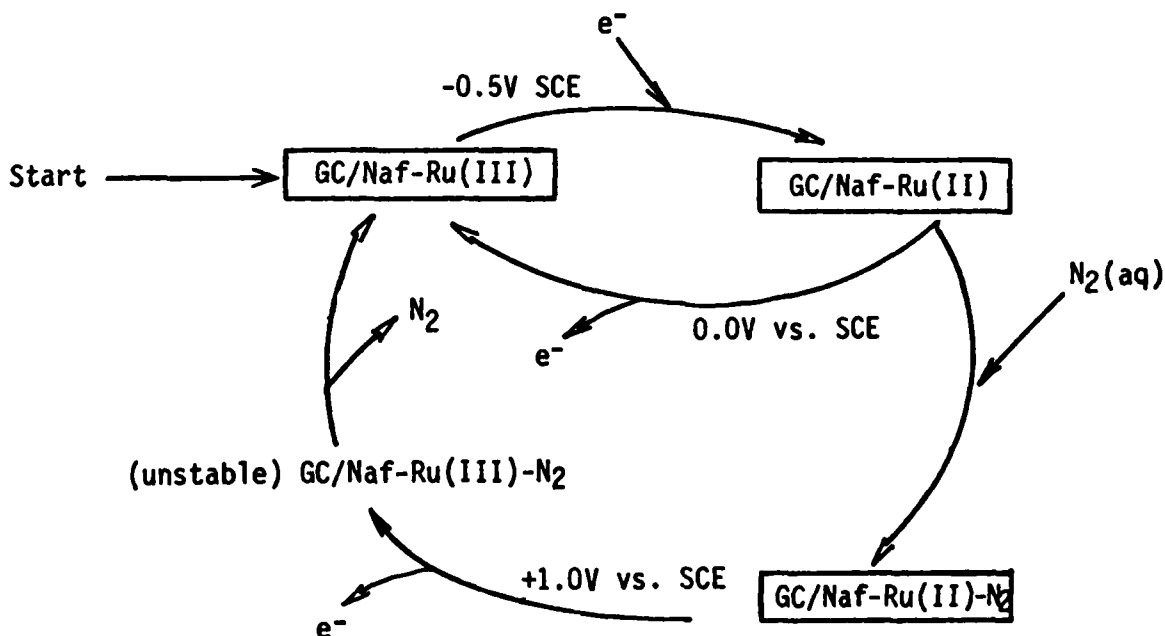
b) Electrolyte composition. The order of stability of the Nafion-bound Ru(III) cation toward electrolyte composition is the following: H₂SO₄ = HCl > NaCl = serum or plasma > K₂SO₄ > KCl. Up to 24 hours of operation of the GC/Naf-Ru electrodes were possible in both physiological saline, pH 7.2 and whole serum or plasma. Air saturation of electrolyte had no effect on the Ru^{+2/+3} electrochemistry, although most experiments were performed in Ar or N₂ saturated electrolyte. The longevity of the Nafion-bound Ru could be increased by the addition of a second layer of Nafion over the Naf/Ru film; however, there was some sacrifice of electrochemical properties, e.g., reversibility and kinetics of electron transfer.

c) Polymer loading of Ru. The initial rate of Ru efflux from Nafion films followed first order kinetics and, as such, was in direct proportion to the initial concentration of Ru within the film. As polymer sites became vacant, or if the polymer sites were not saturated initially, the rate of Ru cation efflux became slower than that predicted by first order kinetics for a dissociation. Thus, lower loadings of Ru appear advantageous for long term use of GC/Naf-Ru electrodes.

4.0 OPERATION OF A GC/Naf-Ru ELECTRODE AS A N₂ SENSING ELECTRODE

The GC/Naf-Ru electrode is activated for N₂ measurement by applying a reducing potential of -0.5V vs. SCE to generate the N₂ complexing Ru(II) cation on the electrode surface. After some time, *t*, at -0.5V to allow N₂ complexation to proceed, the potential of the electrode is slowly ramped to ca. 1.0V vs. SCE to oxidize the residual Ru(II) and the newly formed Ru(II)-N₂ complex. The electrode potential can then be returned to -0.5V to reactivate the Ru(II) cation and initiate a new N₂ complexation reaction sequence, or, switched to open circuit to maintain the Ru in the nonreactive Ru(II) state. Since Ru only reacts with N₂ when it is in the reduced Ru(II) form, the reaction can be started and stopped precisely by controlling the potential of the electrode. Moreover, oxidation of the Ru(II)-N₂ complex causes dissociation of the N₂ and restoration of the Ru(III) cation so that a new cycle of reaction can be initiated at will. Thus reaction rates can be measured for short times, and can be measured repeatedly to follow changes in solution N₂ tension.

The sequence of reactions occurring on the electrode surface are summarized in the following scheme:



Experiments performed under N_2 pressures of 1 to 10 atm demonstrated that the pseudo first-order rate constant, k' , calculated from the change in concentration of Ru(II) during time, t , was proportional to PN_2 . These kinetic experiments are discussed in greater detail in the manuscript (5) appended to this report.

Measurements made under dynamic conditions of changing PN_2 demonstrated that the reaction rate increased when the N_2 pressure over the solution was raised, and decreased when the N_2 pressure was lowered. A typical result, illustrated in Figure 8, suggested that the actual

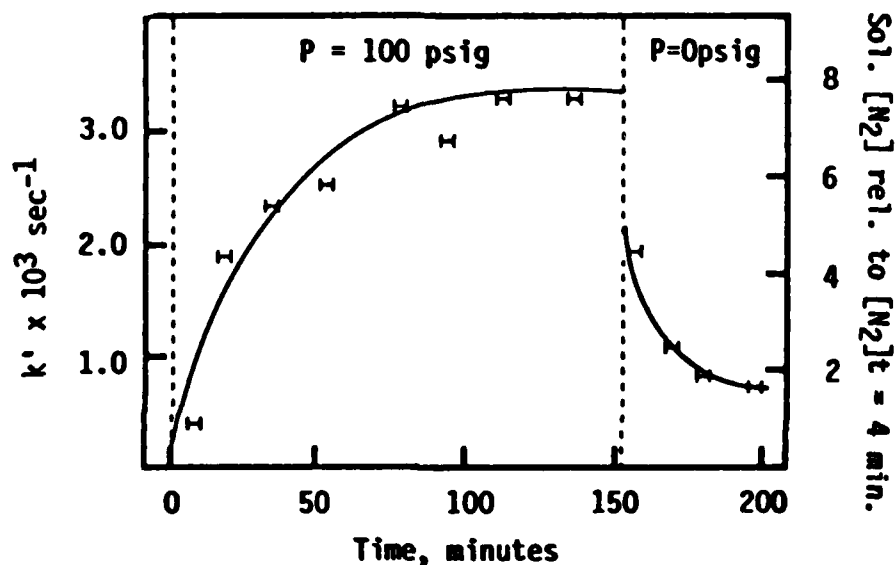


Fig. 8. Variation in reaction rate for Ru(II)- N_2 formation during equilibrium of solution PN_2 . Pseudo first-order rate constant, k' , determined for 4 min reactions of surface Ru(II) with solution N_2 .

concentration of N_2 in solution equilibrated slowly with the N_2 pressure in the gas phase above the solution during both the compression and decompression stages of the experiment. In this experiment, the pressure changes were made as rapidly as possible. Future experiments might employ different compression and decompression schedules, e.g., slow, linear increases and decreases in pressure, or stepwise changes in pressure, and follow the corresponding changes in solution PN_2 with the N_2 sensing electrode.

5.0 CONCLUSIONS AND CONSIDERATIONS FOR FURTHER DEVELOPMENT

It is evident from the results of this study that the unique reactivity of Ru pentaammine, cis-Ru tetraammine and cis-Ru ethylenediamine toward molecular N_2 can be utilized to achieve an electrochemical sensor for quantitating P_{N_2} in solutions. The Ru complexes can be immobilized onto electrode surfaces through ion exchange into an electrode bound cation exchange polymer, Nafion. The surface bound Ru complexes react directly with N_2 in aqueous solution. The reaction between surface Ru complexes and solution N_2 can be followed by electrochemical techniques. At short reaction times, the reaction follows first order kinetics with a rate constant 10 times greater than for the equivalent homogeneous reaction. The electrode kinetics respond to a change in P_{N_2} between 1-8 atm and the electrode sensitivity approaches 1/10 atm.

The N_2 sensing system developed during this program was configured on a glassy carbon electrode. It is probably the first sensor ever which displays potentials for tissue P_{N_2} measurement. With additional development, the sensing system of Nafion-bound Ru cation could be configured for implantation into animal tissues so that in vivo measurements of tissue N_2 tension and exchange under hyperbaric conditions would become feasible. The availability of an implantable N_2 sensor for making direct measurements of the rates of transfer of respiratory gases in different tissue locations would make a significant contribution toward the Navy research programs in hyperbaric medicine, formulation of decompression tables and the prevention of decompression illness.

6.0 REFERENCES

1. A. D. Allen and C. V. Senoff, *Chem. Commun.* 621 (1965).
2. A. D. Allen and J. R. Stevens, *Chem. Commun.* 1147 (1967).
3. M. M. Khan and A. E. Martell in Homogeneous Catalysis by Metal Complexes (New York: Academic Press, 1974), pp. 181-191 and references cited therein.
4. L. S. Robblee and M. A. Parker, Final Report, Contract No. N00014-82-C-0133, "Development of Nitrogen Sensor for Determination of PN_2 in Body Tissues," July 1982.
5. T. J. Lewis and L. S. Robblee, "Development of a N_2 Sensor for Determination of PN_2 in Body Tissue," in Electrochemical Sensors for Biomedical Application, ed. C.K.N. Li (Pennington, New Jersey: The Electrochemical Society, 1986), in press.
6. C. M. Elson, J. Gulens, I. J. Itzkovitch and I. A. Page, *Chem. Commun.* 875 (1970).
7. J. Gulens and J. A. Page, *Electroanal. Chem. and Interfacial Electrochem.* 55:239 (1974).
8. J. D. Buhr and H. Taube, *Inorganic Chem.* 19:2425 (1980).
9. T. Matsubara and P. C. Ford, *Inorganic Chem.* 15:1107 (1976).
10. J. A. Broomhead and L. A. P. Kane-Maguire, *J. Chem. Soc. (A)*, 546 (1967).
11. C. M. Elliot and R. W. Murray, *Anal. Chem.* 48:1247 (1976).
12. N. Oyama and F. C. Anson, *J. Am. Chem. Soc.* 101:3450 (1979).
13. C. R. Martin and K. A. Dollard, *J. Electroanal. Chem.* 159:127 (1983).
14. M. N. Szentirmay and C. R. Martin, *Anal. Chem.* 56:1898 (1984).

APPENDIX

DEVELOPMENT OF A N_2 SENSOR FOR DETERMINATION
OF P_{N_2} IN BODY TISSUE

Manuscript to appear in:

Proceedings Volume, Electrochemical Sensors for
Biomedical Applications, ed. C.K.N. Li,
Electrochemical Society, NJ, 1986

DEVELOPMENT OF A N_2 SENSOR FOR DETERMINATION OF PN_2 IN BODY TISSUE

T. J. Lewis and L. S. Robblee
EIC Laboratories, Inc., 111 Downey Street, Norwood, MA 02062

ABSTRACT

The cationic Ru complexes, $[\text{Ru}(\text{NH}_3)_5\text{H}_2\text{O}]^{3+}$, $[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{3+}$ and $[\text{Ru}(\text{en})_2(\text{H}_2\text{O})_2]^{3+}$ have been immobilized onto electrode surfaces through ion exchange into Nafion polymers. These complexes react directly with dinitrogen in aqueous solution when the ruthenium complexes are reduced to Ru(II). Formation of the nitrogen complexes on electrode surfaces was followed by cyclic voltammetry and the electrode surfaces were characterized by FTIR. At short reaction times, the reaction follows first-order kinetics with a rate constant 10 times greater than for the equivalent homogeneous reaction. The electrode kinetics respond to a change in the PN_2 and the electrode sensitivity approaches 1/10 atm. The minimum electrode response time at elevated PN_2 was 10 seconds.

INTRODUCTION

Decompression sickness is a consequence of bubble formation in tissues supersaturated with an inert gas. A primary goal of formulating decompression schedules for divers and other underwater workers is the avoidance of bubble formation with its accompanying debilitation. A central problem in predicting the onset of bubble formation is incomplete knowledge of inert gas solubility, transport, and exchange in body tissues, particularly above atmospheric pressure (1). Bubbles, once formed, may be detected by ultrasonic means (2-4) or by electrical conductivity measurements in some tissues (5), but this is of little value in predicting or preventing their formation. Clearly, the direct determination of tissue gas tensions and measurements of gas transport and exchange rates between tissue compartments would provide a more accurate basis for formulating safe and efficient decompression regimes.

One way of making in situ measurements of tissue gas tensions is by means of microsensors implanted in the tissue of interest. Sensitive and specific sensors exist which are based upon electrochemical reactions of electroactive species at an electrode surface. For instance, Clarke-type (6) oxygen electrodes use the reduction of molecular oxygen at a cathodically polarized noble metal electrode for PN_2 measurements in vivo (7).

Nitrogen presents a formidable electrochemical detection problem since its oxidation and reduction is not possible within a potential range appropriate for an aqueous environment. For electrochemical detection, dinitrogen must be involved in a preliminary chemical reaction forming an electroactive product.

The results by Allen and Senoff (8) demonstrated that simple ruthenium(II)pentaammine complex of dinitrogen $\{[\text{Ru}(\text{NH}_3)_5\text{N}_2]^{2+}\}$ can be prepared. Taube (9) showed that aquo Ru(II)pentaammine, $[\text{Ru}(\text{NH}_3)_5\text{H}_2\text{O}]^{2+}$, reacts directly with dinitrogen in solution to form $[\text{Ru}(\text{NH}_3)_5\text{N}_2]^{2+}$. Page investigated the electrochemistry of $[\text{Ru}(\text{NH}_3)_5\text{N}_2]^{2+}$ (10) and $[\text{Ru}(\text{NH}_3)_5\text{H}_2\text{O}]^{2+}$ (11). Page (10,11) and Taube (13) studied the kinetics of formation of $[\text{Ru}(\text{NH}_3)_5\text{N}_2]^{2+}$ from $[\text{Ru}(\text{NH}_3)_5\text{H}_2\text{O}]^{2+}$ and N_2 in aqueous solution.

The nitrogen binding chemistry and electrochemistry of bis-aquo ruthenium(II)tetraammine, $[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{3+}$ (12), has been shown to mimic that of aquo ruthenium(II)pentaammine. The complex dinitrogen ruthenium(II)bis-ethylenediaammine aquo, $[\text{Ru}(\text{en})_2(\text{H}_2\text{O})(\text{N}_2)]^{2+}$, has been reported (14).

Our goal has been to develop an electrochemical dinitrogen sensor based on the ruthenium chemistry described above, suitable for *in vivo* monitoring of the partial pressure of dinitrogen. We have immobilized appropriate ruthenium complexes to Nafion membrane coated electrodes and demonstrated that the reactions with nitrogen described for the ruthenium complexes in solution also occur on the surface of electrodes. The rate constant for the formation of dinitrogen complexes of ruthenium is an order of magnitude more rapid when the ruthenium is confined to a surface.

EXPERIMENTAL

Chemicals. $[\text{Ru}(\text{NH}_3)_5\text{Cl}]\text{Cl}_2$, purchased from Strem Chemicals, was used without purification. Nafion 117, purchased from Aldrich Chemicals as a 5 wt/vol% solution in lower aliphatic alcohols and 10% water, was used as received or, in some cases, was diluted with absolute ethanol to 0.5 wt/vol% Nafion. $[\text{Ru}(\text{NH}_3)_4\text{Cl}_2]\text{Cl}$ was synthesized by Dr. S. Pell. $[\text{Ru}(\text{en})_2\text{Cl}_2]\text{Cl}$ was prepared by a modification of a procedure by Broomhead (15). Water was doubly distilled from alkaline KMnO_4 under argon. Salts and acids were ultrapure from commercial sources when available.

Electrode Materials. Glassy carbon electrodes possessing flat, easily polished, circular working surfaces of 0.072 cm^2 were purchased from Bioanalytical Systems. Counter electrodes consisted of platinum mesh or wire coil of substantially larger surface area than the working electrodes. Saturated calomel electrodes (SCE) or sodium saturated calomel electrodes (SSCE) provided potential reference.

Gases. Prepurified nitrogen was used as received. Argon was stripped of residual oxygen by passage through a 0.1M solution of CrCl_2 in 0.5M H_2SO_4 followed by passage through distilled water. Cr(II) was continuously regenerated with Zn/Hg, present in the scrubbing solution.

Equipment. Electrochemical experiments were carried out using an Amel Model 551 potentiostat/galvanostat controlled by a Princeton Applied

Research Corp. Model 175 function generator. Output was recorded on a Bascom-Turner Model 4120 storage X-Y recorder. All electrochemical experiments were conducted in a one compartment electrochemical cell under a controlled atmosphere of argon, nitrogen or air. An IBM IR-38 combined with a Herrick Diffuse Reflectance accessory was used to characterize modified electrode surfaces.

Solutions. Solution electrochemical studies were carried out in a sulfate electrolyte, 0.001M H₂SO₄-0.099M K₂SO₄ pH 3.3 or 0.1M H₂SO₄ pH 1.1, unless otherwise indicated. Electrochemistry of surface modified electrodes was studied in a variety of electrolytes, including 0.1M H₂SO₄, 0.1M HCl, 0.001M H₂SO₄-0.099M K₂SO₄, 0.1M KCl, 0.1M NaCl or phosphate buffered saline (PBS), 0.73M NaCl in 0.1M Sorensen phosphate buffer, pH 7.3.

Surface Modified Electrodes. Nafion was applied to electrode surfaces from alcoholic solution by dip or drip coating. In some instances, the Nafion film was spin-coated onto circular electrodes. Upon solvent evaporation (>½ hr for a 1 µl application), the Nafion was irreversibly adsorbed to the electrode surface. These electrodes are referred to as GC/Naf. Thicknesses of wet Nafion films were calculated using a density of 1.58 g/cm³ (16).

Incorporation of ruthenium cations was carried out by immersing a Nafion-containing electrode in an aqueous solution of the appropriate Ru(III) cation, 10 µM to 10 mM Ru(III), in 0.001-0.1M supporting electrolyte. These electrodes are referred to as GC/Naf-Ru.

Preparation of Nitrogen Complexes of Ruthenium. [Ru(NH₃)₅N₂]²⁺, [Ru(NH₃)₄(H₂O)(N₂)]²⁺ and [Ru(en)₂(H₂O)(N₂)]²⁺ were prepared following a published procedure for the preparation of [Ru(NH₃)₅N₂]²⁺ (9).

Hyperbaric Experiments. Reactions at pressures >1 atmosphere of N₂, air, and Ar were carried out in a Parr Pressure Reactor fitted with electrical feed-throughs for electrode connections. A magnetic stirrer was used in the bulk electrolyte to promote equilibration between the gas and aqueous phase. Before studying electrode kinetics at various PN₂, gas at elevated pressure was allowed to equilibrate with solution for at least 20 minutes before acquiring data. In other experiments, the internal pressure of the bomb was rapidly raised to 8 atmospheres from 1 atmosphere or lowered from 8 atmospheres to 1 atmosphere in less than 15 seconds. Electrode reactions were monitored during gas/solution equilibration. In some instances, inlet gas was passed through the electrolyte solution to insure rapid equilibration between gas at elevated pressure and the solution.

RESULTS AND DISCUSSION

Ruthenium Cations in Nafion Films. Ruthenium cations were incorporated into electrode bound Nafion films through ion exchange. The

ruthenium complexes as the chloride salts, $[\text{Ru}(\text{NH}_3)_5\text{Cl}]\text{Cl}_2$, $[\text{Ru}(\text{NH}_3)_4\text{Cl}_2]\text{Cl}$ and $[\text{Ru}(\text{en})_2\text{Cl}_2]\text{Cl}$, were dissolved in water, in some instances containing the electrolyte, to a final concentration of 10 μM to 10 mM . The Ru cations were incorporated into the GC/Naf electrodes by soaking the electrodes in the Ru containing solutions. Ion exchange of the electroactive Ru cations into the GC/Naf was evaluated by cyclic voltammetry. Formal electrochemical potentials, E° 's, of relevant electroactive Ru complexes in solution and within Nafion membranes are given in Table 1. The E° ' for $[\text{Ru}(\text{NH}_3)_5\text{Cl}]^{2+/1+}$, $[\text{Ru}(\text{NH}_3)_4\text{Cl}_2]^{1+/0}$ and $[\text{Ru}(\text{en})_2\text{Cl}_2]^{1+/0}$ in aqueous solution ranges from -0.19V to 0.23V vs. SCE (11). In the Nafion polymer, E° 's for these ruthenium complexes were shifted 80-100 mV cathodically, consistent with the behavior of $\text{Ru}(\text{NH}_3)_6^{3+/2+}$ in solution and in a Nafion polymer (17). Upon reduction of the GC/Naf-Ru electrodes (Ru = $[\text{Ru}(\text{NH}_3)_5\text{Cl}]^{2+}$, $[\text{Ru}(\text{NH}_3)_4\text{Cl}_2]^{1+}$ or $[\text{Ru}(\text{en})_2\text{Cl}_2]^{1+}$) to generate Ru(II) on the electrode surface, the redox potentials of the complexes shifted anodically as aquo ligands displaced chloro ligands. This is consistent with the behavior of these Ru(II) ions in solution (11).

For GC/Naf electrodes immersed in Ru containing electrolyte solutions, cyclic voltammograms (CVs) were recorded continuously between -0.5V and 0.0V vs. SCE to observe the growth of the wave associated with the $\text{Ru}^{3+} \rightleftharpoons \text{Ru}^{2+}$ interconversions of the reversible ruthenium complexes. Solution concentration of Ru cations were micromolar and cyclic voltammetric signals for these solution species were barely detectable at naked reversible electrodes. Growth of waves associated with the $\text{Ru}^{3+} \rightleftharpoons \text{Ru}^{2+}$ process was rapid, the solution and film reaching equilibrium with respect to Ru cation within minutes. Ion exchange distribution coefficients, k_D ($k_D = [\text{M}^{n+}]_{\text{film}}/[\text{M}^{n+}]_{\text{aq}}$), for these Ru cations were 10^4 , in good agreement with Martin for $\text{Ru}(\text{NH}_3)_6^{3+}$ exchanged into Nafion (17).

The cyclic voltammetry for the Nafion bound $[\text{Ru}(\text{NH}_3)_5\text{H}_2\text{O}]^{3+/2+}$, $[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2]^{3+/2+}$ and $[\text{Ru}(\text{en})_2(\text{H}_2\text{O})_2]^{3+/2+}$ couples in blank electrolyte was consistent with reversible surface-confined reactants for thin (1 μl of 0.5 wt/vol% Nafion) films of Nafion (16). Anodic and cathodic peaks were not or only slightly separated ($\Delta E_p = 0-20$ mV) at low sweep rates ($v \leq 50$ mV/s) and the peak width at half height ($\Delta E_{p1/2}$) was 90 mV. Formal potentials (E° 's) are given in Table 1. The E_p values shifted from $\sim -0.2\text{V}$ vs. SCE in solution to about -0.3V vs. SCE in the Nafion film which is explained by a strong electrostatic interaction between the Ru cations and the Nafion polymer (17). For thick films of Nafion (0.5 μl of 5.0 wt/vol%), behavior suggestive of mass transport limitations controlled the shape of the cyclic voltammograms at $v \leq 20$ mV/s. The E° 's of the redox couples remained at $\sim -0.3\text{V}$ vs. SCE (still shifted from the solution E° ' value), however, $\Delta E_p > 0$ and $\Delta E_{p1/2} > 90$ mV. The peak broadening is attributable to repulsive interactions (19) and spacial distributions of the redox centers within the films (20). The charge transport process has been explained by White (21) as a combination of physical diffusion of the electroactive species and an electron-transfer within the Nafion films.

Apparent diffusion coefficients, D_{app} , for $[Ru(NH_3)_5(H_2O)]^{3+/2+}$ within Nafion membranes were determined from chronocoulometric plots of Q vs. $t^{1/2}$ and from chronoamperometric plots of i vs. $t^{-1/2}$. These plots showed excellent linearity (correlation coefficients always >0.99) over time intervals in which semi-infinite linear diffusion applies ($<1s$). D_{app} for the oxidation of GC/Naf- $[Ru(NH_3)_5(H_2O)]^{2+}$ was 2.1×10^{-9} cm^2/sec .

Contributions to D_{app} are made from electron hopping diffusion (described by an electronic diffusion coefficient, D_{et}) and true molecular or ionic diffusion (described by the ionic diffusion coefficient D_o). Martin (16) and Anson (22) report $Ru(NH_3)_6^{3+/2+}$ diffusion in Nafion is controlled by ionic diffusion. Our value of 2.1×10^{-9} cm^2/sec for $[Ru(NH_3)_5(H_2O)]^{3+/2+}$ is consistent with the value of 3.4×10^{-9} cm^2/sec for $Ru(NH_3)_6^{3+/2+}$ obtained by Martin (16). The diffusion coefficient, D , for $[Ru(NH_3)_5(H_2O)]^{3+/2+}$ in aqueous solution is 6.6×10^{-6} cm^2/sec (11). That the film diffusion coefficient was three orders of magnitude lower than the solution value indicates that the path for diffusion through the Nafion film is convoluted and obstructed, possibly by penetration of chain material. Diffusion may also be inhibited by electrostatic interactions.

Since the Ru cations are electrostatically, not covalently, bound to the Nafion polymer, some loss of Ru can be expected through ion exchange or partition with the solution. The rate of efflux was conveniently monitored electrochemically by using the magnitude of the oxidation or reduction peaks at $\sim -0.3V$ vs SCE as a direct measure of Ru concentration within the polymer. Factors which affected the overall rate of efflux were the following:

(a) Ru cation. No differences were observed between the three Ru compounds $[Ru(NH_3)_5(H_2O)]^{3+}$, $[Ru(NH_3)_4(H_2O)_2]^{3+}$, and $[Ru(en)_2(H_2O)_2]^{3+}$. These Ru(III) cations were more stably bound than the Ru(II) cations, $[Ru(NH_3)_5(H_2O)]^{2+}$, $[Ru(NH_3)_4(H_2O)_2]^{2+}$ and $[Ru(en)_2(H_2O)_2]^{2+}$ due to the greater charge density on the ions.

(b) Electrolyte composition. The order of stability of the Nafion-bound Ru cations toward electrolyte composition was as follows: $H_2SO_4 \sim HCl > NaCl > K_2SO_4 > KCl$. The stability of the Nafion-bound Ru in NaCl electrolyte was increased by the application of a second layer of Nafion over the Naf-Ru film.

(c) Polymer loading of Ru. The initial rate of Ru efflux from Nafion films followed first order kinetics and, as such, was in direct proportion to the initial concentration of Ru within the film. As polymer sites became vacant, or if the polymer sites were not saturated initially, the rate of Ru cation efflux became slower than that predicted by first order kinetics for a dissociation. Apparently, a given Ru cation interacts with more than one ion exchange site within the polymer at low loading. Thus lower polymer loading of Ru is advantageous for long term use of GC/Naf-Ru electrodes.

Reaction of N₂ with GC/Naf-Ru in H₂SO₄ and HCl. The GC/Naf-Ru electrodes prepared as described were evaluated as nitrogen sensing electrodes. Since most solution studies on the direct formation of [Ru(NH₃)₅N₂]²⁺ from [Ru(NH₃)₅H₂O]²⁺ and dissolved N₂ have been performed in electrolyte containing H₂SO₄ (10,13), we used the same electrolyte for initial studies with the GC/Naf-Ru electrodes. The labile Ru(II) species was generated on the electrode surface by poisoning the electrode at -0.5V vs. SCE. The progress of the N₂ complexing reaction was followed by cyclic voltammetry using the decreasing magnitude of the current peak at ~-0.3V vs. SCE for the GC/Naf-[Ru(NH₃)₅H₂O]²⁺ oxidation to indicate consumption of the precursor in forming surface nitrogen complexes. This current peak also decreases due to efflux of Ru(II) from the polymer but the two pathways can be distinguished.

For a given electrode, an initial potential cycle was made from -0.5V to 1.0V back to -0.5V vs. SCE in Ar saturated electrolyte to establish both the quantity of immobilized Ru and the background current in the potential region 0.0V to 1.0V vs. SCE where nitrogen complexes of Ru(II) are expected to undergo oxidation (Figure 1a). The electrode was then poised at -0.5V vs. SCE in N₂ saturated electrolyte for a fixed time, with occasional potential cycles made to 0.0V vs. SCE to observe the Ru^{3+/2+} redox reactions. The current peak at ~-0.3V vs. SCE for the GC/Naf-Ru^{3+/2+} couple progressively decreased with time until it was only a fraction of its initial value (Figure 1b). A subsequent potential cycle to 1.0V vs. SCE revealed two oxidation peaks centered around 0.5V and 0.65V vs. SCE and a single reduction peak at ~-0.3V vs. SCE (Figure 1c). This reduction was less than that observed at the start of the experiment (Figure 1a) due to Ru(II) efflux. An immediate second cycle to 1.0V vs. SCE demonstrated recovery of bound Ru(II) precursor and the absence of oxidation peaks at 0.5V and 0.65V vs. SCE (Figure 1d). The entire experiment was repeated several times with the same sequence of voltammetric responses.

When the sequence of voltammetric measurements was carried out in the absence of N₂, using Ar or He to deaerate the electrolyte, no peaks were observed at 0.5V and 0.65V vs. SCE and the current peaks for the Ru^{3+/2+} couple at ~-0.3V vs. SCE reflected only the change in Ru concentration due to efflux of Ru cations from the polymer.

Exposure of GC/Naf. electrodes to Ru(II)-N₂ complex prepared in solution, followed by cyclic voltammetry measurements in blank electrolyte solution, resulted in voltammetric curves nearly identical to those shown in Figures 1c and 1d.

Infrared spectroscopy was used to characterize the GC/Naf-Ru electrodes. For infrared studies, the surface nitrogen complexes were formed from the reaction of GC/Naf-[Ru(NH₃)₅H₂O]²⁺, GC/Naf-[Ru(NH₃)₄(H₂O)₂]²⁺ or GC/Naf-[Ru(en)₂(H₂O)₂]²⁺ with N₂ at 100 psig in 0.015M HCl. Nitrogen complex formation was monitored by cyclic voltammetry using the decrease in the current peak of the precursor complex as criterion

for the reaction. After 90% decline in the $\text{Ru}^{3+/2+}$ precursor peak, the electrodes were removed from potentiostatic control and dried. An FTIR spectrum of each electrode was acquired using a diffuse reflectance sampling accessory as illustrated for ruthenium pentaamine in Figure 2, curve 1. Peaks attributed to the surface nitrogen complexes of ruthenium pentaamine, tetraamine, and bis(ethylenediamine) occurred between 2100 and 2150 cm^{-1} (23).

Each electrode was examined by cyclic voltammetry between -0.5V and 0.8V vs. SCE after infrared characterization. Current peaks at 0.5V and 0.65V vs. SCE attributable to the oxidation of the surface nitrogen monomers and dimers were observed. The electrodes were then potentiostated at 0.8V vs. SCE until all surface nitrogen species were decomposed, as determined by cyclic voltammetry for each electrode. The infrared spectra of each electrode were again obtained and are shown for ruthenium pentaamine in Figure 2, curve 2. Peaks attributable to the surface nitrogen complexes of ruthenium pentaamine, tetraamine, and bis(ethylenediamine) disappeared with the disappearance of the cyclic voltammetric waves associated with the surface ruthenium nitrogen complexes.

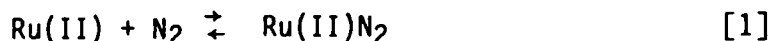
Literature values for the peak positions of the N-N stretching frequencies of the nitrogen complexes prepared in aqueous solution and the stretching frequencies of the Nafion immobilized nitrogen complexes are reported in Table 2. Infrared spectra of GC/Naf-Ru(II) electrodes not exposed to N_2 , and GC/Naf-Ru(III) exposed to dinitrogen only in the Ru(III) state, showed no absorption attributable to an N-N stretch.

Kinetics of GC/Naf-Ru Reaction with N_2 . The rate of formation of surface ruthenium-nitrogen complexes was determined in an electrochemical pressure vessel with N_2 pressures ranging from 0 to 80 psig. After 20 minutes equilibration at any given N_2 pressure, a GC/Naf-Ru electrode was poised at -0.5V vs. SCE (to convert and maintain the bound Ru in the labile Ru(II) state) for various times, e.g., 1, 2, 4, 8, and 16 minutes. After these time intervals, two complete potential cycles were made to 1.0V and back to -0.5V vs. SCE. The quantity of Ru(II) complex present on the electrode as a function of time at each P_{N_2} was determined from the areas under the current peaks at $\sim -0.3\text{V}$ vs. SCE during the first and second cycles of each set of measurements. The relevant areas are illustrated in Figure 1c and 1d. Making two measurements for each time interval provided an internal calibration of the system and minimized errors arising from Ru(III) efflux from the polymer.

Plots of $\ln[\text{Ru(II)}]_0/[\text{Ru(II)}]_t$ vs. time were linear for each Ru(II) cation and P_{N_2} tested, Figure 3. The slopes of these plots provided values for the pseudo first-order rate constants (k') for each reaction. The logarithms of $t^{1/2}$ vs. $\log \text{P}_{\text{N}_2}$ are shown in Figure 4. The linearity and slope of these log-log plots indicates the formation of N_2 complexes obeyed first order kinetics with respect to $[\text{N}_2]$, similar to

previous observations in solution studies (24). Deviation from linearity at the highest pressure suggests that the actual solution P_{N_2} was lower than indicated by gas pressure readings, or that contribution from another process such as bis-ruthenium dinitrogen dimer formation (10,11,13) was becoming important.

The reaction occurring on the GC/Naf. electrode surface is:



This reaction proceeds at the rate

$$d[\text{Ru(II)N}_2]/dt = k[\text{Ru(II)}][\text{N}_2] \text{ moles cm}^{-2}\text{sec}^{-1} \quad [2]$$

Since $[\text{N}_2]$ is essentially constant throughout the reaction, compared to $[\text{Ru(II)}]$, the reaction follows pseudo first-order kinetics and the rate expression can be written as:

$$d[\text{Ru(II)N}_2]/dt = k'[\text{Ru(II)}] \text{ moles cm}^{-2}\text{sec}^{-1} \quad [3]$$

where $k' = k[\text{N}_2] \text{ sec}^{-1}$.

The pseudo first-order kinetics of reaction [1] permits the experimental determination of k' from the ratio of initial $[\text{Ru(II)}]$ to $[\text{Ru(II)}]$ at t seconds according to the relationship:

$$k' = (1/t) \ln[\text{Ru(II)}]_0/[\text{Ru(II)}]_t$$

Pseudo first-order rate constants for the formation of GC/Naf.- Ru(II)N_2 for each Ru complex at various P_{N_2} are given in Table 3. The rate constants for formation of the nitrogen complexes on electrode surface is 10 times greater than those reported for the solution reaction (11). The threshold of our detection corresponds to ~2% conversion of GC/Naf.- Ru(II) according to equation [1]. At the high pressure limit in this study, a GC/Naf.- Ru(II) electrode has a response time of 10 sec and electrode sensitivity approaches 1/10 atm N_2 .

Figure 5 illustrates the changes in k' under dynamic conditions of changing N_2 pressure in the electrochemical cell. At the start of the experiment, the pressure in the cell was increased in a single step to 100 psig (in <20 sec), held at that level for 152 minutes and then quickly released to bring the cell back to 0 psig (in <15 sec). Each value of k' was determined for a 4 minute reaction of GC/Naf.- $[\text{Ru}(\text{NH}_3)_5\text{H}_2\text{O}]^{2+}$ with solution N_2 at different times after raising or lowering the N_2 pressure. Each value of k' is shown on the plot as a bar in order to indicate the time of the GC/Naf.- $[\text{Ru}(\text{NH}_3)_5\text{H}_2\text{O}]^{2+} + \text{N}_2$ reaction in relation to the overall time of the experiment. The k' values increased with time after the N_2 pressure was raised and became constant after about 100 minutes. When the pressure was released, k' values decreased rapidly. The observed changes in the pseudo first-order rate constants, k' , during the pressurization and depressurization of the cell with N_2 are presumed to be due to the changing concentration of dissolved N_2 in solution.

The final value of k' , determined 1 hour after depressurization to 1 atm N_2 was greater than the initial value of k' at 1 atm N_2 (determined from a standard deviation of several electrodes at 1 atm N_2). This result is similar to that of Ross (25) who studied gas sensing electrodes possessing a porous membrane and internal electrolyte. The time response of such gas sensing electrodes was reported to be dependent upon the electrode geometry, membrane properties and electrolyte. In general it was determined that outgassing of a membrane containing internal electrolyte was considerably slower than the reverse process.

CONCLUSIONS

The cationic Ru complexes, $[Ru(NH_3)_5H_2O]^{3+}$, $[Ru(NH_3)_4(H_2O)_2]^{3+}$ and $[Ru(en)_2(H_2O)_2]^{3+}$ can be immobilized onto electrode surfaces through ion exchange into an electrode bound cation exchange polymer, Nafion. The ruthenium complexes are tightly bound within the Nafion polymer even in 0.1M electrolyte solution and are more tightly bound to Nafion than to classic cation exchange polymers. The cyclic voltammetric responses of GC/Naf-Ru(II)/(III) electrodes are ideal-Nernstian and mimic what is known for Nafion immobilized $Ru(NH_3)_6^{3+/2+}$. The surface bound Ru(II) complexes react directly with N_2 in aqueous solution. At short reaction times the reaction follows first-order kinetics, with a rate constant 10 times greater than for the equivalent homogeneous reaction. The electrode kinetics respond to a change in the P_{N_2} and the electrode sensitivity approaches 1/10 atm. The minimum electrode response time at elevated P_{N_2} was ~10 sec. The reaction between GC/Naf-Ru(II)/(III) electrodes and N_2 are most well behaved in dilute acid solution where efflux of the Ru from the electrode surface is slow.

Acknowledgments: We wish to thank Peter Marren for technical assistance and the Office of Naval Research for financial support.

REFERENCES

1. Weathersby, P. K., Homer, L. D., Undersea Biomedical Research, 7, 227 (1980).
2. Walder, D. N., Evans, A. and Hempleman, H. V., Lancet, 1:897 (1968).
3. MacKay, R. S., Rubissow, G., in Underwater Physiology, Proc. Fourth Symposium on Underwater Physiology, ed. C. J. Lambertsen, Academic Press, New York, 1971, p. 151.
4. Spencer, M. P., and Smith, M. T., Undersea Biomedical Research, 6(suppl.):16 1979).
5. Hills, B. A., *ibid*, p. 115.
6. Clark, L. C., Jr., Trans. Amer. Soc. Art. Internal Organs, 2, 41 (1956).

- 7a. Clark, L. C., Jr., *J. Appl. Physiol.*, 13(I), 85 (1958).
- b. Kreuzer, F. and Kimmich, M. P., in *Measurement of Oxygen*, eds. H. Degn, I. Balslev and R. Brook, Elsevier, Amsterdam, 1976, p. 123 and references cited therein.
- c. Caspers, H. and Speckmann, E. J., *Epilepsia*, 13, 699 (1972).
- d. Leniger-Follert, E. and Lubbers, D. W., *Pflugers Arch.*, 366, 39 (1976).
8. Allen A. D. and Senoff, C. V., *J. Chem. Soc., Chem. Commun.*, 621, (1965).
9. Harrison, D. E. and Taube, H., *J. Am. Chem. Soc.*, 89, 5706 (1967).
10. Itzkovitch, I. J. and Page, J. A., *Can. J. Chem.*, 46, 2743 (1968).
11. Elson, L. M., Itzkovitch, I. J., McKenney, J., and Page, J. A., *Can. J. Chem.*, 53, 2922 (1975).
12. Elson, C. M., Itzkovitch, I. J., and Page, J. A., *Can. J. Chem.*, 48, 639 (1976).
13. Armor, J. N. and Taube, H., *J. Am. Chem. Soc.*, 92, 6171 (1970).
14. Kane-Maguire, L. A. P., Sheridan, P. S., Basolo, F. and Pearson, R. G., *J. Am. Chem. Soc.*, 90, 5295 (1969).
15. Broomhead, J. A., Kane-Maguire, L. A. P., *J. Chem. Soc., (A)*, 546 (1967).
16. Martin, C. R., Dollard, K. A., *J. Electroanal. Chem.*, 159, 127 (1983).
17. Szentirmay, M. N., Martin, C. R., *Anal. Chem.*, 56, 1898 (1984).
18. Peerce, P. J., Bard, A. J., *J. Electroanal. Chem.*, 114, 89 (1980).
- 19a. Lenhard, J. R., Murray, R. W., *J. Am. Chem. Soc.*, 100, 7870 (1978).
- b. Kno, K., Moses, P. R., Lenhard, J. R., Green, D. C., Murray, R. W., *Anal. Chem.*, 51, 745 (1979).
- c. Abruna, H. D., Meyer, T. J., Murray, R. W., *Inorg. Chem.*, 112, 3233 (1979).
20. Kaufman, F. B., Engler, E. M., *J. Am. Chem. Soc.*, 701, 547 (1979).
21. White, H. S., Leddy, J., Bard, A. J., *J. Am. Chem. Soc.*, 104, 4811 (1982).
22. Buttry, D. A. and Anson, F. C., *J. Electroanal. Chem.*, 130, 333 (1981).

23. Allen, A. D., Boftomky \bar{r} ., Harris, R. O., Rensalu, V. P., Senoff, C. V., J. Am. Chem. Soc., 89, 5595 (1967).
24. Robblee, L. S., manuscript in preparation.
25. Ross, J. W., Riseman, J. H., Krueger, J. A., Pure and Applied Chemistry, 39, 473 (1973).

TABLE 1
FORMAL REDUCTION POTENTIALS
OF RUTHENIUM COMPLEXES

	E° (V vs. SCE)†	
	Aqueous Solution	Nafion Film
$\text{Ru}(\text{NH}_3)_6^{3+}$	-0.19 (11)	-0.29 (17)
$\text{Ru}(\text{NH}_3)_5\text{Cl}_2^+$	-0.21 (11)	-0.34
$\text{Ru}(\text{NH}_3)_5(\text{H}_2\text{O})^{3+}$	-0.17 (11)	-0.26
$\text{Cis-Ru}(\text{NH}_3)_4\text{Cl}_2^+$	-0.32 (11)	-
$\text{Cis-Ru}(\text{NH}_3)_4(\text{H}_2\text{O})\text{Cl}_2^+$	-0.21 (11)	-
$\text{Cis-Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2^{3+}$	-0.15 (11)	-0.25
$\text{Cis-Ru}(\text{en})_2\text{Cl}_2^+$	-0.30 §	-
$\text{Cis-Ru}(\text{en})_2(\text{H}_2\text{O})_2^{3+}$	-0.14	-0.24

TABLE 2
STRETCHING FREQUENCIES OF NITROGEN
COMPLEXES OF RUTHENIUM

	cm^{-1} †
$[\text{Ru}(\text{NH}_3)_5\text{N}_2]\text{Cl}_2$	2105 (23)
$[\text{Ru}(\text{NH}_3)_5\text{N}_2](\text{PF}_6)_2$	2167 (23)
$\text{cis-}[\text{Ru}(\text{en})_2(\text{N}_2)_2][\text{B}(\text{C}_6\text{H}_5)_4]$	2220, 2190 (14)
$\text{cis-}[\text{Ru}(\text{en})_2(\text{H}_2\text{O})\text{N}_2][\text{B}(\text{C}_6\text{H}_5)_4]_2$	2130 (14)
GC/Naf.- $[\text{Ru}(\text{NH}_3)_5\text{N}_2]\text{Cl}_2$	2133
GC/Naf.- $[\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})\text{N}_2]\text{Cl}_2$	2083
GC/Naf.- $[\text{Ru}(\text{en})_2(\text{H}_2\text{O})\text{N}_2]\text{Cl}_2$	2085

† Numbers in parentheses refer to reference from which the data were obtained. All other values were determined by the authors.
§ $(E_p)_a$ not observed; value is $(E_p)_c + 0.03\text{V}$.

TABLE 3
PSEUDO FIRST-ORDER RATE CONSTANTS FOR NITROGEN
COMPLEX FORMATION IN NAFION POLYMERS

	P_{N_2} (atm)	k' $\text{sec}^{-1} \times 10^4$
$\text{Ru}(\text{NH}_3)_5(\text{H}_2\text{O})^{2+} + \text{N}_2$	1.0	2.3
	1.7	4.0
	2.3	6.1
	3.7	10.0
	6.3	11.2
$\text{Ru}(\text{NH}_3)_4(\text{H}_2\text{O})_2^{2+} + \text{N}_2$	1.0	3.9
	1.7	5.5
	2.3	8.0
	3.7	11.1
	6.3	20.0
$\text{Ru}(\text{en})_2(\text{H}_2\text{O})_2^{2+} + \text{N}_2$	1.0	2.5
	1.7	3.6
	2.3	5.6
	3.7	7.4
	6.3	11.0

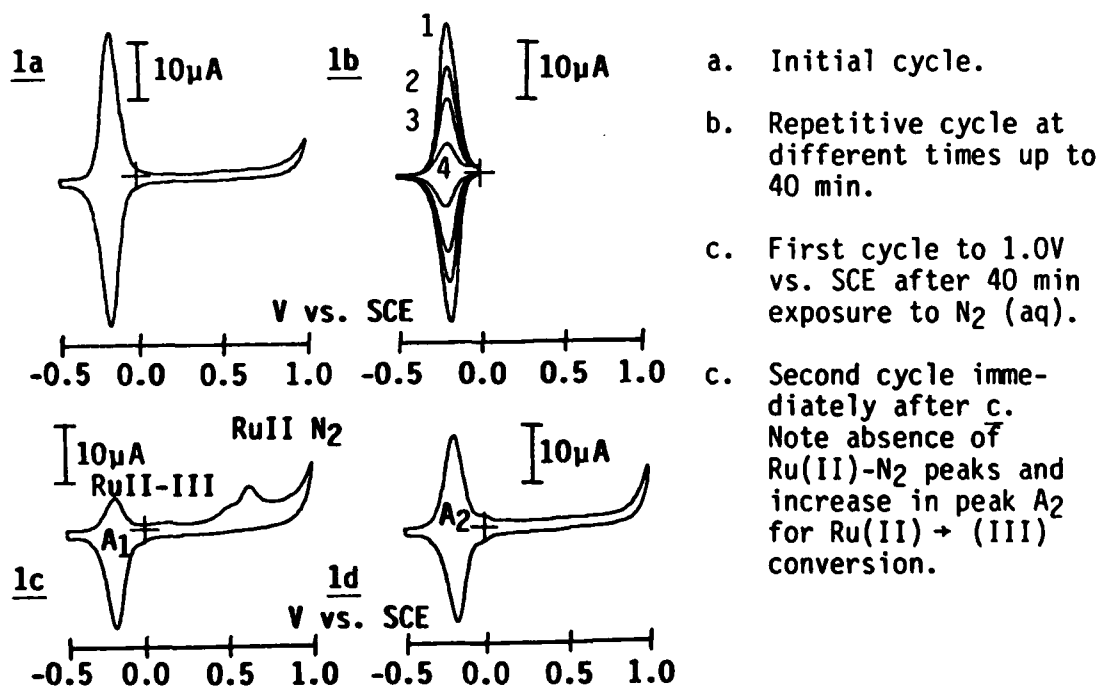


Fig. 1. Cyclic voltammetry of GC/Naf.-Ru electrode in 0.1M H_2SO_4 ; N_2 saturated; sweep rate, 50 mV sec^{-1} .

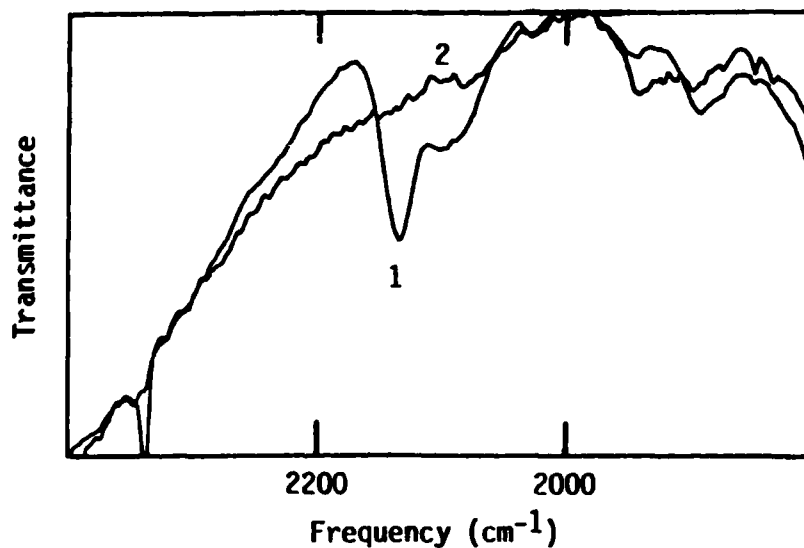


Fig. 2. The FTIR spectra of (1) GC/Naf.- $[\text{Ru}(\text{NH}_3)_5\text{H}_2\text{O}]^{2+}$ upon reaction with N_2 and (2) after electrochemical oxidation at 0.8V vs. SCE of electrode from (1).

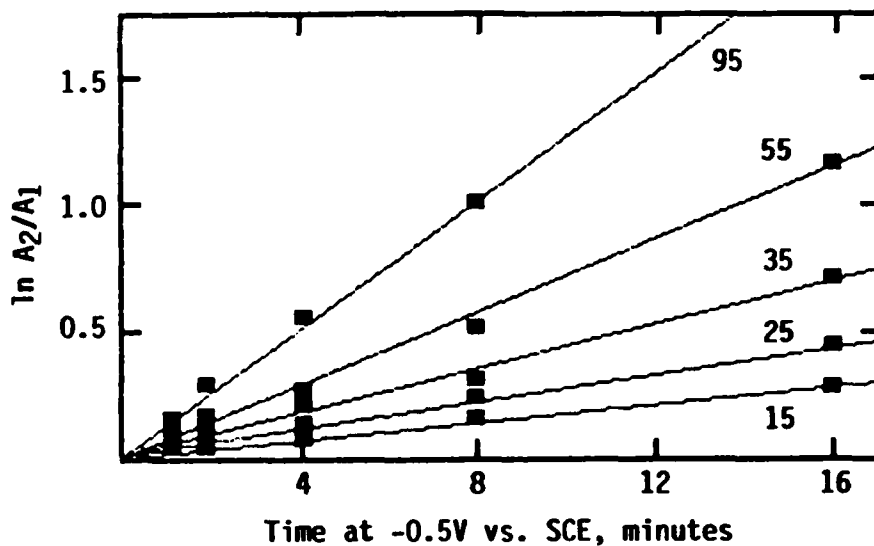


Fig. 3. Rates of reaction with N_2 at different pressures (psi as indicated on graph) for a GC/Naf.-Ru electrode in 0.1M H_2SO_4 . See Figures 1c and 1d for definition of A_2/A_1 .

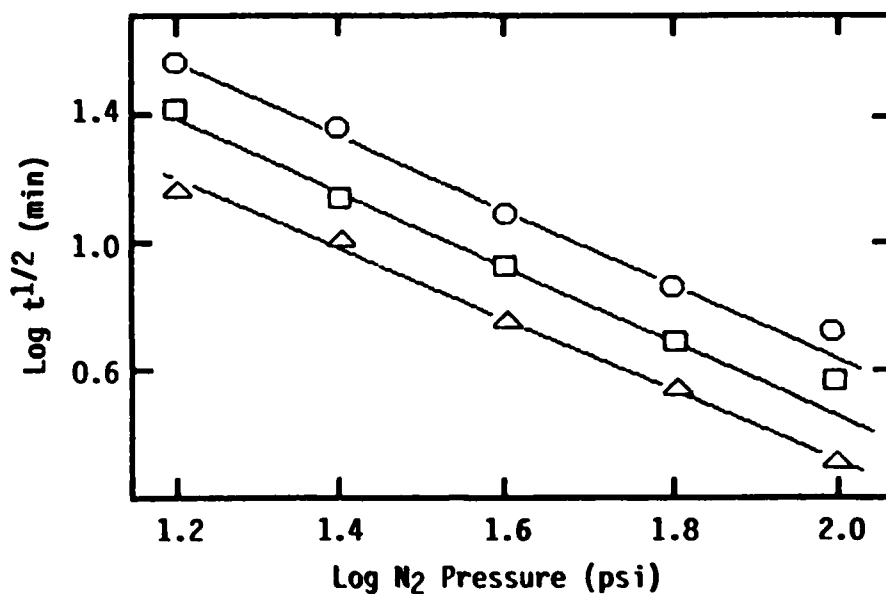


Fig. 4. Rate of reaction with N₂ in 0.1M H₂SO₄ of GC/Naf.-Ru electrodes where Ru is: ○ Ru(en)₂; □ RuN₅; △ RuN₄.

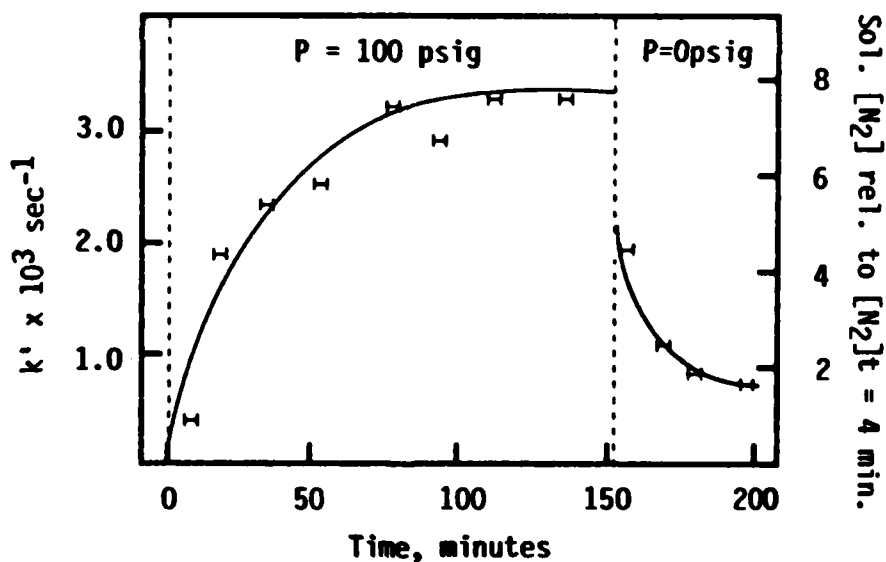


Fig. 5. Variation in reaction rate for Ru(II)-N₂ formation during equilibration of solution P_{N₂}. Pseudo first-order rate constant, k', determined for 4 min. reactions of surface Ru(II) with solution N₂.

OFFICE OF NAVAL RESEARCH
PHYSIOLOGY PROGRAM
DISTRIBUTION LIST FOR TECHNICAL REPORTS

Number of Copies

12

Administrator
Defense Technical Information Center
Bldg. 5, Cameron Station
Alexandria, VA 22314

6

Director
Naval Research Laboratory
Washington, D.C. 20375
Attn: Technical Information Division
(Code 2027)

2

Physiology Program (Code 441NP)
Office of Naval Research
800 North Quincy Street
Arlington, VA 22217

2

Franklin G. Hempel
Scientific Officer
Office of Naval Research
800 North Quincy Street
Arlington, VA 22217

One copy to each of the following:

Administrative Contracting Officer
Code FACC-C2
DCASMA, Boston
495 Summer Street
Boston, MA 02210

Assistant Chief for Technology
Office of Naval Research (Code 200)
800 North Quincy Street
Arlington, VA 22217

Commanding Officer
Naval Medical Research and
Development Command
Naval Medical Command
Bethesda, MD 20814

Office of Naval Research Western
Regional Office
1030 East Green Street
Pasadena, CA 91106

Technical Reference Library
Naval Medical Research Institute
Naval Medical Command
Bethesda, MD 20814

Dr. A. A. Salfkosky
Scientific Advisor, Commandant of
Marine Corps (Code RD-1)
Washington, D.C. 20380

Director
Science and Technology Division
Library of Congress
Washington, D.C. 20450

Commanding Officer
Naval Health Research Center
P.O. Box 85122
San Diego, CA 92138

Director
Air University Library
Maxwell AFB, AL 36112
Attn: AUL-8110

Director
National Library of Medicine
8600 Wisconsin Avenue
Bethesda, MD 20014

Director
Life Sciences Division
Office of Scientific Research
U.S. Air Force
Bolling AFB - Building 410
Washington, D.C. 20332

Human Performance Program
Naval Medical Research and
Development Command (Code 44)
Naval Medical Command
Bethesda, MD 20814

Library
Naval Submarine Medical Research Lab
Naval Submarine Base, New London
Box 434
Groton, CT 06340

Library
Naval Ocean Systems Center
San Diego, CA 92152

Office of Naval Research Scientific
Liaison Group
American Embassy - Room A-407
APO San Francisco, CA 96503

Librarian
Naval Aerospace Medical Institute
Building 1953, Code 012
Pensacola, FL 32508
Attn: Mrs. Ruth Rogers

Commanding Officer
Naval Air Development Center
Warminster, PA 18974
Attn: Life Sciences Research Group

Dr. Robert Campbell
Army Research Office
Research Triangle Park, NC 17709

Office of Naval Research
495 Summer Street
Boston, MA 02210

Commanding Officer and Director
Naval Training Device Center
Orlando, FL 32813
Attn: Technical Library

Commandant
U.S. Air Force School of Aerospace
Medicine
Brooks Air Force Base, TX 78235
Attn: Aeromedical Library (SMSDL)

Library
U.S. Army Aeromedical Research Unit
Fort Rucker, AL 30360

END

10286

DTIC