THE DESIGN AND EVALUATION OF

ION SELECTIVE ELECTRODES

AS SENSORS FOR AUTOMATED ANALYSIS

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ABSTRACT

In this work the design preparation and evaluation of several ion selective electrodes are described. Electrodes have been prepared using both solid state and immobilized liquid ion-exchangers and a novel lead selective electrode, based on an immobilized antibiotic, has been developed. Applications of these electrodes were studied, with special attention to their use as endpoint sensors in the automated techniques of potentiometric titrations. These techniques, utilizing the generation of concentration gradients, include a new variant of the gradient titration known as the 'single cup gradient titration'.

Ion selective electrodes are shown to be effective sensors for automated titrations. They can provide sharp, well defined endpoints either directly or when electronic differentiation of the electrode signal is used to compensate for the greater response time of some immobilized membranes.

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INTRODUCTION

Ion selective electrode potentiometry has undoubtedly been one of the most rapidly expanding areas of analytical chemistry in recent years. This state of affairs was assisted somewhat by some of the over optimistic claims in early work regarding electrode 'specificity'. Fortunately this initial confusion has largely disappeared and the technique seems to be entering a period where a more realistic assessment of the true value of these devices is being made. Despite their limited selectivity in many cases, ion selective electrodes do provide a valuable extension to the range of instrumental analytical techniques and have found applications in routine analysis, chemical studies, biomedical screening and industrial process control.

Early studies on glass membranes have shown that ion exchange is the operative mechanism of electrode response (1,2) and the development of this idea has led to the discovery of many new electrodes selective to a wide variety of species. The ionic activity of almost any species can be sensed; anions, cations, gases, enzymes and substrates as well as complex organic compounds, the problem being not in the sensing of the species but in achieving sufficient selectivity. Even this latter limitation is not significant if the analysis is coupled with the relevant separation chemistry, e.g. ion exchange, chromatography etc. Ion selective electrodes (known widely as ISE's) are inexpensive. The low cost of sensors and ancilliary equipment has been one of the main reasons for

their extensive use in routine laboratory applications. An added advantage is that analysis is non destructive, which is especially important in 'on-line' use of ISE's for industrial process control, pollution monitoring and clinical-biomedical screening. The simplest of ion selective electrode techniques will give an accurate analysis with the minimum of effort and if more advanced techniques are used, ion selective electrodes can provide a high level of precision in both manual and automated methods of analysis. A survey of the literature will show a vast range of applications, many of which are given in a comprehensive review by Koryta (3) and the biannual reviews of Buck (4,5,6).

SECTION I

The Development and Principles of

Ion Selective Electrodes

CHAPTER 1

The Development of Ion Selective Electrodes.

When a thin membrane of some materials separates two solutions, a potential difference is formed across the membrane. In 1906 Cremer (7) found that the potential produced across a glass membrane was dependent upon the pH of the solutions on either side. This led to the easy and rapid determination of pH by means of the 'glass electrode'. In 1922 Hughes (8) first described what became known as the 'sodium error' of glass electrodes. This interference was systematically investigated in an attempt to reduce it, and after 1947 it was realized that the 'error' could be utilized to produce electrodes sensitive to alkali metal ions (9,10,11). By altering the composition of the glass membrane, electrodes sensitive to many different ions have been produced (H^+, Li^+, Na^+, K^+, Cs^+, Mg^{2+}, Ca^{2+}, etc.) and although a specific response to a selected ion could not be obtained, selective electrodes were made. An example of these is the sodium electrode, which is 10^3 times more sensitive to Na^+ than it is to K^+, but which is also sensitive to most other monovalent ions.

In attempts to make ion specific electrodes many different materials have been used as membranes, for example single crystals, clays, ion exchange resins, collodion, cellophane and precipitates impregnated into various inert materials. In 1936 Tendeloo devised a method for the potentiometric determination of calcium concentrations in solutions (12) using a thin plate of fluorite (CaF_2) as a

membrane, sealed in the end of a glass tube. Although Tendeloo quoted a molar concentration range of three decades and mentioned the use of the electrode for measuring calcium in milk (9), both Anderson (13) and Kolthoff and Sanders (14) found that the electrode failed.

After Nicolskii (1) introduced the concept of ion-exchange to explain the mechanism of the glass electrode, work commenced on the theories for ion-exchange electrodes and the application of ion-exchangers to selective electrodes. Kolthoff and Sanders (14) examined the electrode properties of fused silver chloride and silver bromide pellets. In an indirect way they showed the ion selective properties of these membranes, but their results were overlooked by analytical chemists. The early work carried out in the field of clay membranes has been reviewed by Marshall (15). The surfaces of the clay membrane are capable of exchanging ions with those in solution, but as the clays are anionic in nature, the membranes prepared from them are useful for the determination of cationic activities only. Electrodes sensitive to both monovalent and divalent cations were produced but their concentration range was small and their selectivity for ions of the same charge, and frequently for ions of different charges, was poor.

Ion exchange resins have the advantage over clay membranes in that anion sensitive as well as cation sensitive forms can be produced, although electrodes reversible to anions other than those incorporated in the membrane are not generally known. Most workers in this

field have used commercially available ion-exchangers, e.g. Ionac C-200, Amberlite IR-100, Amberlite IR-120-K etc., although a few have prepared their own (16). The ion-exchange resins, like clay membranes, show little selectivity for different ions, and are generally only cation or anion sensitive. An additional problem arises when the electrodes are used in a high concentration of an interfering ion. The membrane sensitive to chloride for example, when used in a strong bromide solution, may change its form and become a bromide sensitive membrane, an effect which can cause serious errors in potentiometric titrations. Various ideas and methods to produce selective ion-exchange resin membrane electrodes have been attempted. Active groups have been incorporated into the resins (17,18), and these have been cast, pressed into pellets and made up with polystyrene binders. The results of this work were disappointing even though some analytical work, e.g. the total cationic activities in milk (19), was carried out. Pungor, using resins with a silicone rubber binder (20), incorporated complexonites but found no increase in selectivity.

Since extensive studies of ion distribution had already been carried out on liquid ion exchangers and it was known that they possessed a high selectivity for specific ions, they seemed to be suitable exchangers for membrane electrodes (21). The next stage in electrode development was therefore the use of hydrophobic liquids containing dissolved ion exchangers (22,23). The first

practicable liquid membrane electrode was produced in 1966 by Ross (24) and was selective towards calcium ions. The exchanger used was a calcium salt of didecyl phosphoric acid dissolved in di-n-octyl phenyl phosphonate, which was supported by cellulose dialysis tubing. Electrodes selective to nitrate (25), perchlorate (26), and potassium (27) were soon developed and many applications were found. The major drawbacks with this type of system were that the electrode used a large amount of expensive ion-exchanger and was mechanically weak as well as being difficult to prepare and short lived. These factors were overcome by immobilization of the ion-exchanger in an inert matrix to produce a solid heterogeneous membrane. The most versatile method of immobilization is the incorporation of the liquid exchanger in a P.V.C. (poly vinyl-chloride) matrix, providing a rugged rubbery membrane such as those developed by Moody and Thomas (28).

The solid heterogeneous membrane electrodes were initiated by the work of Wylie and Patnode in 1950 (29), although much progress has been made since then. It was considered that the incorporation of a slightly soluble salt might cause ion selectivity, and barium sulphate was a likely candidate. It was incorporated into cellophane membranes by Hirsch-Ayalon (30), paraffin by Fisher and Babcock (31), and silicone rubber by Pungor (32). Like many electrodes of this type the membranes were not very selective, though they could respond to barium and sulphate as well as hydrogen ions. Of the precipitate impregnated

membranes, reviewed by Covington (33), the most successful have been those based on silicone rubber as the matrix material. Electrodes with good selectivity towards iodide, chloride, bromide and sulphide have been made using precipitates of the silver salts (34,35,36).

Possibly the ultimate heterogeneous membrane is one in which the membrane is composed entirely of the ion exchanger in a mechanically stable form, usually a compressed disc. Numerous electrodes have been described (3) including those selective towards, lead, cadmium and copper (37,38). These electrodes, consisting of a membrane made of pressed mixtures of silver sulphide and the sulphides of lead, cadmium or copper (II) respectively, respond to silver ion activity, which is controlled by the sulphide ion activity, which in turn depends on the activity of the divalent metal in solution (26). The membrane will thus respond to the metal M^{2+} in solution, and be selective, provided that the solubility product of this ion with sulphide ion is substantially larger than the solubility product of silver sulphide. An electrode responsive to sulphate, based on the lead electrode with incorporated lead sulphate, has also been described (39). A unique case of a homogeneous membrane is the fluoride ion selective electrode (40). The sensitive membrane consisting of a single crystal of lanthanum fluoride, possibly doped with another rare earth such as europium, has very few interferences. The fluoride specificity results from the small size of the LaF_2^+ sites which are present in the pure

crystal, while ionic transport takes place via defects in the crystal structure.

Most electrodes in common use have an internal filling solution but in some applications, for example when an electrode is used inverted, this is inconvenient as electrical contact within the electrode may be broken. To overcome this difficulty solid contacts between the membrane and connecting wires have been developed. Contact with the surface of silver based membranes can most easily be made with silver loaded epoxy resins (38) although this is not successful for the iodide membrane. Solid contact versions of the immobilized liquid ion exchangers have been made by coating platinum wires (41) and graphite (42) with the exchanger. A similar concept has resulted in the development of the 'Selectrode' by Růžička (43,44,45). The electrode consists of a hydrophobitised graphite cylinder which is impregnated with the electroactive species. Contact is made by direct connection with a steel wire. These developments have been useful but with the exception of a silver contact to a silver based membrane, the standard potentials of the electrodes (E_o) may be liable to drift (46,76). The single or mixed crystal membranes have the advantages of being easy to produce, inexpensive, robust and having a long working life. Unfortunately the range of ions for which such membranes can be made selective is limited.

Membrane potentials were initially studied as models for biological systems. In the past few years this process has been reversed, one of the most significant

advances in the field of ion selective electrodes stemming from the observation that neutral macrocyclic antibiotics induced ion permeation in mitochondria (47). Using this knowledge, Simon and co-workers have investigated a wide range of antibiotics for the development of 'neutral carrier' membrane electrodes (48-51), the most promising of which was valinomycin (51,52). Valinomycin dissolved in an appropriate solvent (hexane or diphenyl ether) was used as the liquid ion exchanger in a conventional liquid membrane electrode. The selectivity of this electrode is of the order of 5000:1 for potassium over sodium which makes it a potent tool for analytical and biochemical measurements.

The related but structurally simpler cyclic polyethers, many of which have been synthesised by Pederson (53), have been found to produce cation-selective membrane electrodes with good selectivity towards individual ions. However at present none of these can compete with the selectivity provided by the valinomycin electrode. Although most of the currently available neutral carrier membrane electrodes are responsive to univalent cations, some synthesised exchangers have been produced to respond to divalent cations such as barium and calcium. The barium electrode (54,55) is based on a polyethylene glycol derivative which contains twelve ethylene oxide units per mole of Ba^{2+} and two moles of tetraphenyl-borate ion. A selectivity ratio of 10^4 for barium over calcium and several other cations has been claimed. The calcium electrode (56)

uses non-cyclic compounds and can be tailored to be responsive towards barium or sodium. This tailoring of exchanging complexes seems to be an almost limitless means of extending the range of electrodes available to the analyst. In addition, the utilization of enzyme coupled reactions to produce species for which electrodes are already available, has brought the analysis of many medically important organic compounds within the capability of ion selective electrodes (57,58,3).

It is probable that a more detailed study of the selectivity/structure relationship will help in the production of 'ion-selective molecules', which with the aid of enzyme reactions will produce electrodes with selectivities superior to those obtained so far.

CHAPTER 2

The Theory and Performance of Selective Membrane Electrodes

2.1 Membrane Potential

2.2. The Solid State Membrane

2.2.1. Sensitivity and Detection Limit

2.2.2. Selectivity

2.2.3. Time Response

2.3. Liquid Membranes with Electrically Charged Ligands

2.3.1. Sensitivity and Detection Limit

2.3.2. Selectivity

2.3.3. Time Response

2.4. Liquid Membranes with Electrically Neutral Ligands

2.4.1. Sensitivity and Detection Limit

2.4.2. Selectivity

2.4.3. Time Response

2.5. Immobilized Matrix Liquid Membrane Electrodes

2.5.1. Sensitivity and Detection Limit

2.5.2. Selectivity

2.5.3. Time Response

2.6. Dependence of Potential on Flow Rate

2.6.1. Streaming Potential

2.6.2. Non-streaming Potential Dependence on Flow Rate

2.1. Membrane Potential.

In 1909 Haber and Klemensiewiez (59) proved that the potential generated across a glass membrane varied with the hydrogen ion activity in the solutions contacting it, according to the now familiar Nernst equation.

$$E = Eo + {}^{RT}/_{F} \ln (H^{+}) \qquad 2.1.-1$$

E = potential across membrane, Eo = standard electrode potential, R = gas constant, T = absolute temperature, F = the Faraday's Constant, (H^{+}) = the ionic activity of the hydrogen ion.

After the 'sodium error' of pH electrodes (8) was discovered and investigated, it was realized that the Donnan (60) and Teorell-Meyer-Sievers theories (61,62) of membrane potentials (assuming porous or semipermeable membranes) could no longer explain the electrode properties. The work of Lengyel and Blum (63) had already shown that the potential is formed only in the surface layer of the glass membrane, and led Nicolskii and Tolmacheva (1,64) to develop the concept of an ion exchange reaction for potential formation. They derived the equation:

$$E = E_o + \frac{RT}{nF} \ln (a_i + K_{ij}^{Pot} a_j) \qquad 2.1.-2$$

where a_i is the ion being determined, a_j is the interfering ion and K_{ij}^{Pot} is the selectivity coefficient (preference of sensor for ion j in relation to ion i). A universal integral equation has been derived to explain the steady state E.M.F. of an electrochemical cell containing any type of ion selective membrane electrode (65).

2.2. The Solid State Membrane

2.2.1. Sensitivity and Detection Limit.

Precipitate-exchange phenomena of various materials have been utilized to prepare homogeneous and heterogeneous solid state membranes. As the electrode configurations differ only in mechanical and not in electrochemical respects, and the most important of these are the electrodes based on the silver ion, this section will be limited to membranes prepared from silver salts (Ag_zX). The equation for the membrane potential is therefore:

$$E = E^{o}_{Ag} + \frac{RT}{F} \ln a^{1}_{Ag} \qquad 2.2.1.-1$$

Using the solubility product : $L_{Ag_zX} = (a^{1}_{Ag})^{z} a^{1}_{X}$

the equation can be rewritten:

$$E = (E^{o}_{Ag} + \frac{RT}{zF} \ln L_{Ag_zX}) - \frac{RT}{ZF} \ln a^{1}_{x}$$

$$= E^{o}_{x} - \frac{RT}{ZF} \ln a^{1}_{x} \qquad 2.2.1.-2$$

which holds for sample solutions containing as anions only X^{z-} with an activity a^{1}_{x} at the boundary contacting the membrane.

The lower detection limit of the membrane, as described by Simon (66), is dictated by the membrane material and related to the minimal activity a^{1}_{Ag} which is given either by dissolution of the membrane (solubility) or the silver defect concentration at the membrane surface. When the defect activity, α , is assumed constant for a given silver compound, the following equation describes the deviation in ionic activities between the boundary

(a^1) and bulk (a) of sample solution:

$$a^1_{Ag} - a_{Ag} = Z\,(a^1_X - a_X) + \alpha \qquad 2.2.1.-3$$

Using this relationship the E.M.F. responses of the cells for different membrane materials has been described.

Membrane Material AgX, $L_{AgX} \ll \alpha^2$ (eg AgCl).

The equation derived for a cationic response is:

$$E = E^o_{Ag} + \frac{RT}{F} \ln \frac{a_{Ag} + \sqrt{a_{Ag}^2 + 4\,L_{AgX}}}{2} \qquad 2.2.1.-4$$

and for anionic response:

$$E = E^o_X - \frac{RT}{F} \ln \frac{a_X + \sqrt{a_X^2 - 4\,L_{AgX}}}{2} \qquad 2.2.1.-5$$

This symmetrical response is in agreement with practical observation, as the Nernstian response only holds for $a_{Ag} \gg \sqrt{L_{AgX}}$. The detection limit is therefore set by the level of silver or halide ion released by dissolution of the membrane.

Membrane Material Ag_zX ; $\alpha^{z+1} \gg z.L_{Ag_zX}$

e.g. AgI, Ag_2S

In this case the equation derived for cationic response is:

$$E = E^o_{Ag} + {}^{RT}/_{F} \ln (a_{Ag} + \alpha) \qquad 2.2.1.-6$$

Thus the Nernstian response is only obtained when $a_{Ag} \gg \alpha$ and the detection limit is set by the level of α. The anionic response is more complex, a sudden increase in the E.M.F. with decreasing activity a_x is predicted at $a_x \approx {}^{\alpha}/_{z}$, and it may become unsteady with super-Nernstian response. At low anion levels the equation simplifies to:

$$E = E^o_{Ag} + {}^{RT}/_{F} \ln (\alpha - z\,a_x) \qquad 2.2.1.-7$$

and the detection limit is again controlled by the defect activity α.

2.2.2. Selectivity

Silver halide or halide/sulphide membranes (AgX) may be used to determine the anions (Y^{z-}) which form sparingly soluble salts, Ag_zY, with Ag^+. When these anions occur with X^- they will constitute an interference.

In the case of a solution containing only Y^{z-}, at a high activity level, a layer of Ag_zY (solubility L_{Ag_zY}) will completely cover the membrane and the potential of this electrode is then given by the equation:

$$E = E^o_Y - \frac{RT}{ZF} \ln a_Y \qquad 2.2.2.\text{-}1$$

As the selectivity coefficient (K^{pot}_{xy}) is defined by:

$$E = E^o_x - \frac{RT}{F} \ln (K^{pot}_{xy}\ a_Y^{\,1/z}) \qquad 2.2.2.\text{-}2$$

it can be described in terms of the solubility products of the silver salts: $K^{pot}_{xy} = {}^{L}_{AgX} / {}^{L}_{Ag_zY}{}^{1/z}$ 2.2.2.-3

With solutions such as Br^- (X) and SCN^- (Y), using an AgBr membrane, equation (2.2.2.-1) will describe the potential until the Br^- activity increases to a level given by $a_x \frac{L_{AgX}}{(L_{Ag_zY})^{1/z}} (a_Y)^{1/z}$ at which point the Ag_zY layer will be removed and the membrane will respond to bromide only. If the surface of the membrane consists of more than one halide, for example when using mixed chloride and bromide solutions, then the potential is always given by the equation:

$$E = E^o - \frac{RT}{F} \ln (a_x + K^{pot}_{xy}\ a_y) \qquad 2.2.2.\text{-}4$$

This treatment indicates that the selectivity of a solid state membrane depends only on the solubility products of the silver-interferent salts, and has been extended to derive equations for the response of silver

based membranes towards complexing ligands (66).

2.2.3. Time Response

Although few detailed investigations have been made in this area, the response time of an ion selective electrode is of great importance, especially in its application to continuous monitoring. An electrode which requires hours to come to a steady potential or to achieve a new stable reading after a change in ion activity, would be of little use in a real time analytical situation. Most responses quoted are of the 'static' process, the time for an electrode to reach an equilibrium potential after it has been placed in a solution of ions to which it is sensitive. This time includes the formation of an electrical double layer as well as the establishment of the ion exchange equilibrium. The 'dynamic' time response includes only the latter and is due to the change in ion concentration in the same solution. It must also be noted that whereas the dynamic response times refer to a flowing or stirred solution, the static response times may be obtained in stirred or unstirred solutions. In the latter case the response time may depend on the shape of the membrane as this can affect the double layer formation.

As the most important applications of ion selective electrodes are in the field of automated and continuous analysis, the dynamic time response is of more interest. The problem of a suitable definition of response time has hampered the comparison of results for

a long time, but the quantity t_{95} has recently been proposed (67). This parameter has been defined as the time required for the electrode to achieve 95% of its final equilibrium potential. The values of t_{95} for solid state membranes are sufficiently low to enable their use as continuous sensors. The silver halide electrodes have also been shown to obey first order kinetics in the response to a concentration change (68) and the response times for this type of electrode are given in table 2.2.3.-1.

Table 2.2.3.-1 (From ref. 68)

Electrode	Chloride	Bromide	Iodide
Response Time (in sec) t_{95} $X10^3$	370	200	50
1st. Order Rate Constant	3.46	6.52	26

2.3. Liquid Membranes with Electrically Charged Ligands.

2.3.1. Sensitivity and Detection Limit

Liquid membranes of this type contain a charge bearing ligand or site 'S' which is not soluble in the solutions in contact with it. The membranes are assumed to be permeable only to the counter ions of this site, e.g. 'I'. In contrast to solid ion-exchangers the active sites are free to move within the membrane and as the solvents used are usually of low dielectric constant the association of the species I^+ and S^- is expected within the membrane phase (*) according to the equation (2.3.1.-1)

to result in a neutral ion pair, IS_*,

$$I^+_* + S^-_* \underset{\leftarrow}{\overset{KIS_*}{\rightleftharpoons}} IS_* \qquad 2.3.1.-1$$

which is freely mobile in the membrane.

The liquid membrane can be represented diagramatically by figure (2.3.1.-1).

SOLUTION (')	MEMBRANE (*)	SOLUTION (")
$I^{+'}$ ⟷	I^+_* ⟷	$I^{+"}$
	⟷ S^-_* ⟶	
$X^{-'}$ ⟶		⟵ $X^{-"}$
	⟷ IS_* ⟶	
$J^{+'}$ (interferent)	$J^+_* : JS_*$	($J^{+"}$ interferent)

Figure 2.3.1.-1 from ref. (69). The Liquid Membrane

For such a system the membrane potential is given by (70):-

$$E = \frac{RT}{F}\left\{(1-\tau)\ln\left(\frac{a'_i + \left[\frac{(u^*_j + u^*_s)k_j}{(u^*_i + u^*_s)k_i}\right]a'_j}{a''_i \left[\frac{(u^*_j + u^*_s)k_j}{(u^*_i + u^*_s)k_i}\right]a''_j}\right) + \tau\ln\left(\frac{a'_i + \left[\frac{u^*_{js}}{u^*_{is}}\right]K_{ij}\cdot a'_j}{a'_i + \left[\frac{u^*_{js}}{u^*_{is}}\right]K_{ij}\cdot a''_j}\right)\right\} \qquad 2.3.1.-2$$

$$0 \leqslant \tau \leqslant 1$$

The detection limit of liquid membranes is governed by the solubility of the ions I^+ in the membrane. When the solution contains less I^+ than the membrane there will be a tendency for the ion (and site) to be released from the membrane and go into solution. The amount of ion released into the solution boundary will depend on the

partition coefficient between the sample, and membrane solvents as well as the formation constant of the IS_* complex. The detection limit, for most liquid membranes is of the order of 10^{-5} molar.

2.3.2. Selectivity

The selectivity parameter in the first term of equation (2.3.1.-2) is $(U_{J*} + U_{S*})\ k_J / (U_{I*} + U_{S*})\ k_I$. Thus it depends on both the partition coefficient ratio of the dissociated species (k_J/k_I), and the ratio of the mobilities of the dissociated ion pairs, $(U_{J*} + U_{S*}) / (U_{I*} + U_{S*})$. Both of these terms are controlled mainly by the properties of the solvent. When the value of γ approaches zero (indicating that the ion exchanger is negligibly mobile) this selectivity parameter is controlling and reduces to $(U_{J*}\ k_J / U_{I*}\ k_I)$. Thus increased selectivity can be achieved by using a solvent which differentiates between the analyte and interferent.

The selectivity parameter in the second term in equation (2.3.1.-2), $(U_{JS_*} / U_{IS_*})\ K_{IJ}$, is determined by the product of the mobility ratio of the neutral pairs and the ion exchange equilibrium constant of the reaction:-

$$J^{+'} + IS_* \rightleftharpoons JS_* + I^{+'} \qquad 2.3.2.\text{-}1$$

This term, therefore, depends on the properties of both the solvent and the ion exchanger and when the exchanger is considerably mobile in the membrane ($\gamma \rightarrow 1$), it is the controlling selectivity term. The selectivity parameters have been summarised in table (2.3.2.-1).

Table 2.3.2:-1 From Ref. 3.

Ion exchanger Characteristics	K_{IJ}^{pot}
Strongly dissociated associates in the membrane	$U_J k_J / U_I k_I$
Negligibly dissociated associates in the membrane	$\frac{U_J + U_S}{U_I + U_S} \cdot \frac{k_J}{k_I}$
Ion exchanger negligibly mobile ($\tau \to 0$)	$\frac{U_J}{U_I} \cdot \frac{k_J}{k_I}$
Ion exchanger considerably mobile ($\tau \to 1$)	$\frac{U_{JS}}{U_{IS}} \cdot K_{IJ}$

Summary of selectivity parameters for liquid exchange electrodes with charged ligand sites.

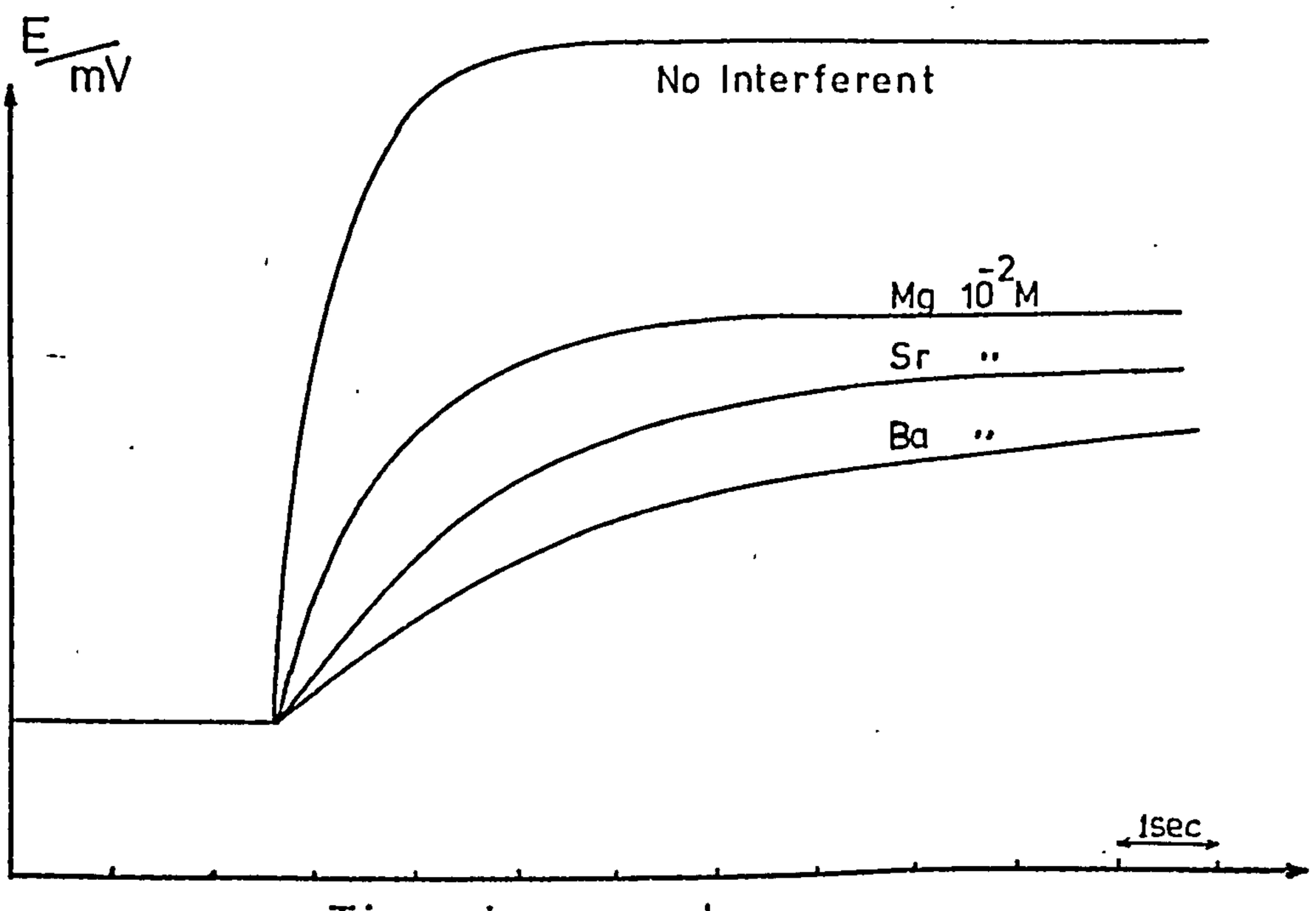
2.3.3. Time Response

Liquid ion-exchanger membrane electrodes are generally more sluggish than the solid state sensors, and their response character can deteriorate rapidly with age. The static response times for membranes such as those responsive to nitrate and calcium are generally quoted as 'a few seconds' (71), the time being dependent on the ionic concentration. Detailed work on the dynamic time response of the calcium electrode provides a t_{95} of 2.3 $\pm$ 0.2 seconds in pure solutions of calcium salts (67,68). The presence of interfering ions in the solution can increase this time by a factor of ten, due to their influence on the equilibrium:-

$$2M_J^{n+} + nCaS_2(*) \rightleftharpoons K \rightleftharpoons nCa^{2+} + 2M_JS_n(*) \qquad 2.3.3.\text{-}1$$

An increase in response time will occur if the dissociation of $M_JS_n(*)$ in the membrane, or the exchange of M_J^{n+} between the membrane and solution becomes rate limiting when the concentration of Ca^{2+} is changed (67). The response time will also be changed if the selectivity coefficient is favourable for the interferent, as it will then occupy a larger proportion of the available exchange sites. Typical results of interferents on the time response of the calcium electrode are shown in figure 2.3.3.-1

Figure 2.3.3.-1 From Ref. (68)



Comparison of the effects of different interferents on the response of the calcium electrode to a change from 10^{-4} to 10^{-3}M

2.4. Liquid Membranes with Electrically Neutral Membranes, 'Neutral Carrier Membrane Electrodes'.

2.4.1. Sensitivity and Detection Limit.

The neutral carrier membrane electrode contains an electrically neutral ligand, S, which is in equilibrium with an electrolyte solution containing cation I^+, and anion X^-. The following reaction is assumed to occur:-

$$I^+ + X^- + S_* \rightleftharpoons IS_*^+ + X_*^- \qquad 2.4.1.\text{-}1$$

Making several assumptions, Eisenman and coworkers (70) derived the following equation for the membrane potential, at zero current, in mixtures of two ions I^+ and J^+ :-

$$E = \frac{RT}{F} \ln \left\{ \frac{a'\ I^+ (U_{JS*}/U_{IS*})(K_J/K_I)\, a'_J}{a''\ I^+ (U_{JS*}/U_{IS*})(K_J/K_I)\, a''_J} \right\} \qquad 2.4.1.\text{-}2$$

where K_I and K_J are the equilibrium constants of the salt extraction reactions ; (2.4.1.-3), which measure

$$I^+ + X^- + S_* \overset{K_I}{\rightleftharpoons} IS_*^+ + X_*^-$$
$$J^+ + X^- + S_* \overset{K_J}{\rightleftharpoons} JS_*^+ + X_*^- \qquad 2.4.1.\text{-}3$$

the extent to which the neutral molecules, S, can extract the salt I^+X^- or J^+X^- from the aqueous to the organic solvent phase. The membrane can be represented schematically by figure 2.4.1.-I (70).

The detection limit is reached when the level of I^+ in the solution becomes lower than the level of I^+ in exchange equilibrium at the surface of the membrane,($\sim 10^{-5}$ M). If this is the case the membranes will respond to the I^+ that has been released from the IS_*^+ complex and returned to the aqueous phase.

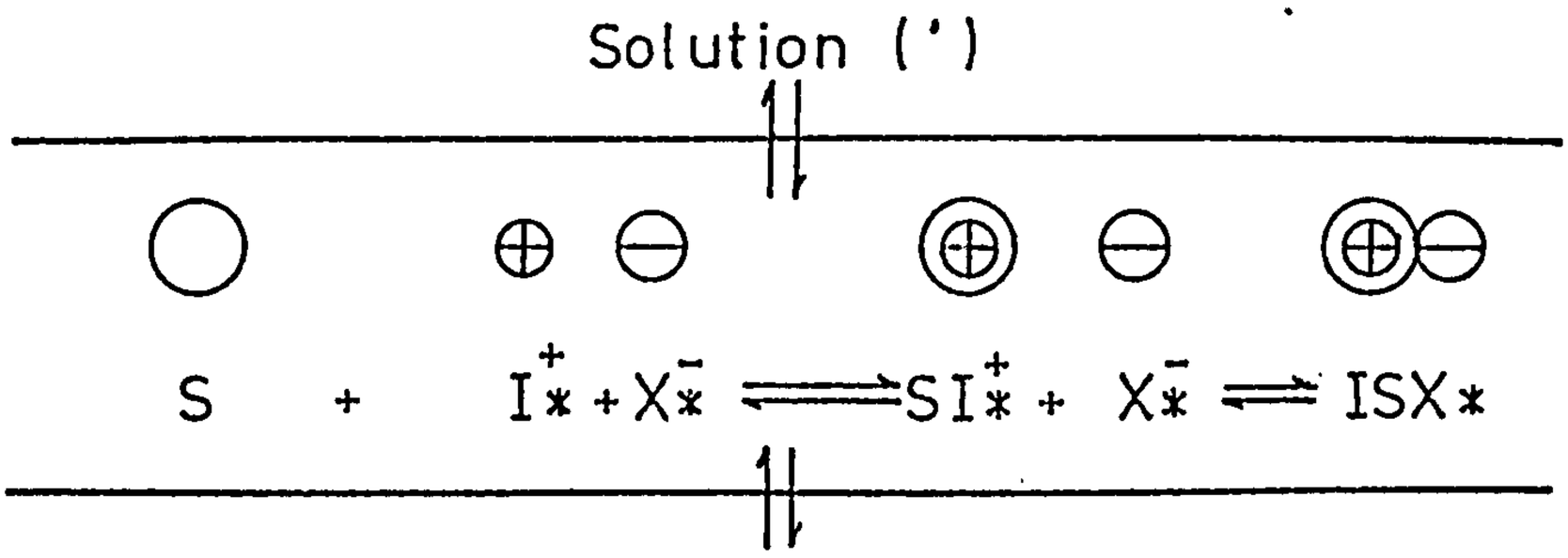


Figure 2.4.1-I
A Schematic Diagram of a Neutral Carrier Membrane Electrode.

2.4.2. Selectivity.

Although equation (2.4.1.-2) is similar to those derived for solid ion-exchange membranes, the physical meaning of the parameters are different. Usually, in neutral carrier membranes, the mobilities of the complexed cations appear to remain constant,i.e. independent of the individual ion. In such cases the selectivity ratio reduces to the term K_J/K_I, the ratio of the equilibrium constants of the exchange reactions. One of the major factors in neutral carrier selectivity is the size of the selected ion, but many other details are important. A comprehensive discussion of the reasons for the high selectivity of ligands such as valinomycin, based on the calculated free energy of transfer of a cation from an aqueous solution to the ligand in the organic phase has been given by Simon (72).

2.4.3. Time Response.

The results obtained for the potassium (valinomycin) electrode are similar to those for the

calcium electrode. The static time response being about 60 seconds, (71), and the dynamic time response (t_{95}) being 3.3 seconds (68). The main difference between the time response of the calcium and potassium electrodes being the latter's independence from interference effects.

2.5. Immobilized-matrix Liquid Membrane Electrodes.

2.5.1. Sensitivity and Detection Limit.

The main property of liquid membrane electrodes which makes them so useful as ion sensors is that their exchanging sites are mobile, enabling their full selectivity to be realised. Rechnitz (73) has reported that the mobile charge carriers (sites, ions, or ion site pairs) are of molecular size and hence mobility of the exchange sites would not be affected by immobilization of the bulk membrane phase, provided that the bulk phase is permeable to the microscopic charge carriers. The fundamental advantages of liquid membranes can therefore be retained with some improvement in the mechanical and physical characteristics of the electrode.

The potential generated by the immobilized exchanger is the same as that for the corresponding liquid electrode but the detection limit is usually lowered due to a reduction in the leaching out of exchanger and complex.

2.5.2. Selectivity.

The selectivity of the immobilized membrane should be the same as for the liquid membrane, although some small changes may be apparent due to the changes in mobility within

the matrix. In practice the selectivity of the calcium electrode over magnesium is slightly improved by immobilization.

2.5.3. Time Response.

The response times of P.V.C. immobilized electrodes have recently been surveyed by Moody and Thomas (71) and the effects of immobilization on dynamic responses have been summarized by Ryan (68):

(1) The rates of the two reactions governing potential formation (surface ion exchange and diffusion within the membrane) are affected differently by immobilization. The surface exchange is quicker and the diffusion rate is slower, thus the two stages of response are apparent even in single ion solutions (contrasting with the liquid membranes) as can be seen in figure 2.5.3.-I.

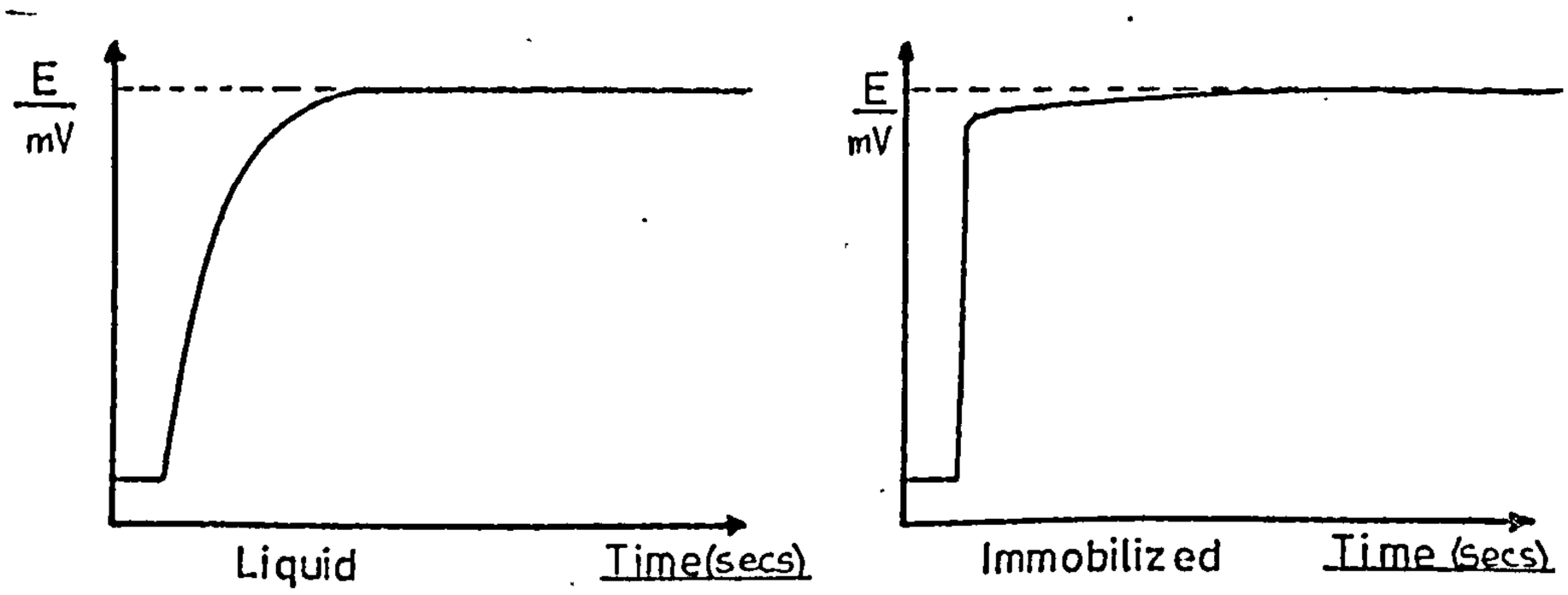


Figure 2.5.3.-I

Time Response Profiles of Liquid and Immobilized Calcium Electrodes.

(2) For the calcium P.V.C. membrane, in pure calcium solutions, the membrane diffusion is rapid and the value of t_{95} is decreased (t_{95} = 1.4 sec.). In the

case of the potassium P.V.C. membrane, however, the valinomycin complex is considerably less mobile and t_{95} increased.

(3) A consequence of point (1) is that the values of t_{95} become slightly concentration dependent, which was not the case for the liquid membrane.

(4) The effects of interfering ions on the response time is apparent at a lower activity level and thus the tolerable analytical level of most interferents is reduced.

2.6. Dependence of Potential on Flow Rate.

When ion selective electrodes are used in continuous flow applications, using proportioning pumps, potential oscillations are frequently observed. Although these oscillations have been attributed to liquid junction potential changes at the reference electrode and to static electricity effects, a recent paper has shown that they are, in part at least, due to streaming potential changes (74).

2.6.1. Streaming Potential.

When a liquid is forced through a narrow capillary by a pressure gradient, a potential difference E_S is set up between the ends of the capillary. This potential is called the streaming potential and is described by the Helmholtz-Smoluchowski equation (2.6.1.-1):

$$E_S = \frac{D \cdot \zeta}{4\pi\eta} \cdot \frac{\Delta P}{\lambda} \qquad 2.6.1.-1$$

where D = dielectric constant, ζ = electrokinetic potential (volt), η = viscosity (poise), ΔP = pressure gradient (dyne cm^{-2}), λ = conductivity ($ohm^{-1} \cdot cm^{-1}$).

If one assumes that the flow in the tube is laminar:

$$\Delta P = \frac{8d\ \eta\ l}{\pi\ r^4} \qquad 2.6.1.-2$$

d = flow rate (ml/sec), l = length of capillary, or length of the tube connecting the electrodes (cm), r = radius of the capillary (cm). Substitution of (2.6.1.-2) in (2.6.1.-1) gives:-

$$Es = 2\ \frac{dDl\zeta}{\pi^2 r^4 \lambda} \qquad 2.6.1.-3$$

where divided by 9.10^{11} , Es is in volts.

From equation (2.6.1.-3) it can be seen that any variation in flow rate changes the streaming potential which will be seen by the reference and sensor electrodes. Some variation in the flow rate will occur due to the ageing of pump tubing, but the major fluctuations are caused by the removal of pressure from the bars of the peristaltic pump. This latter effect can change the flow rate from slightly negative to double the average flow rate of the solution. The pulsing can be considerably reduced by using air bubble segmentation of the solution, thus introducing a cushioning effect, and also by using a flow restrictor (a length of narrow bore tubing) which acts as a pulse suppressor thus reducing Es by a factor of up to 100. The most effective way of reducing the streaming potential is to increase the conductivity of the solution (λ). As Es is inversely proportional to the conductivity, increasing the salt concentration of the solution to a level of 5×10^{-2} M or above, will reduce the value of the streaming potential to an acceptable level.

The cell designed for use in differential titrations may also be used to reduce the effects of pulsing. The electrode membranes are held parallel to each other in the same plane as the solution flow, Figure (2.6.1.-I). With this design the value of 'l' in equation (2.6.1.-3) is small, as the streaming potential across each membrane will be the same (assuming laminar flow) and Es will be ignored when the potential difference between the two membranes is measured.

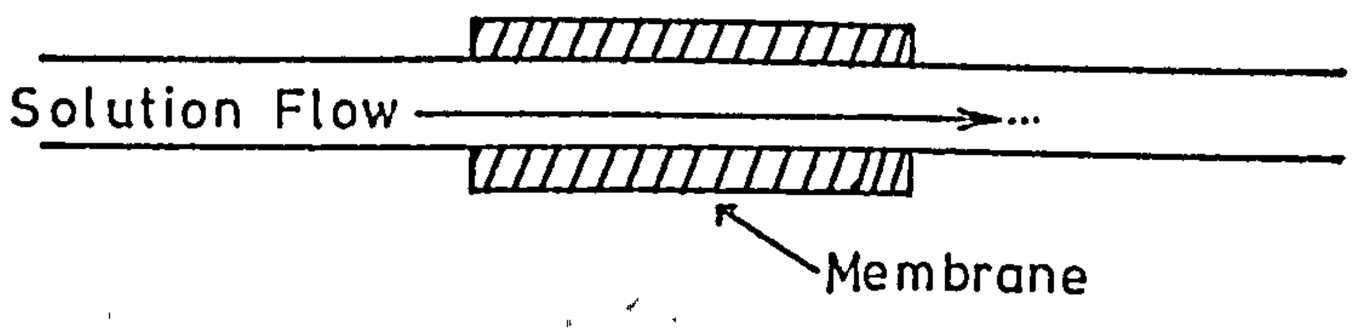


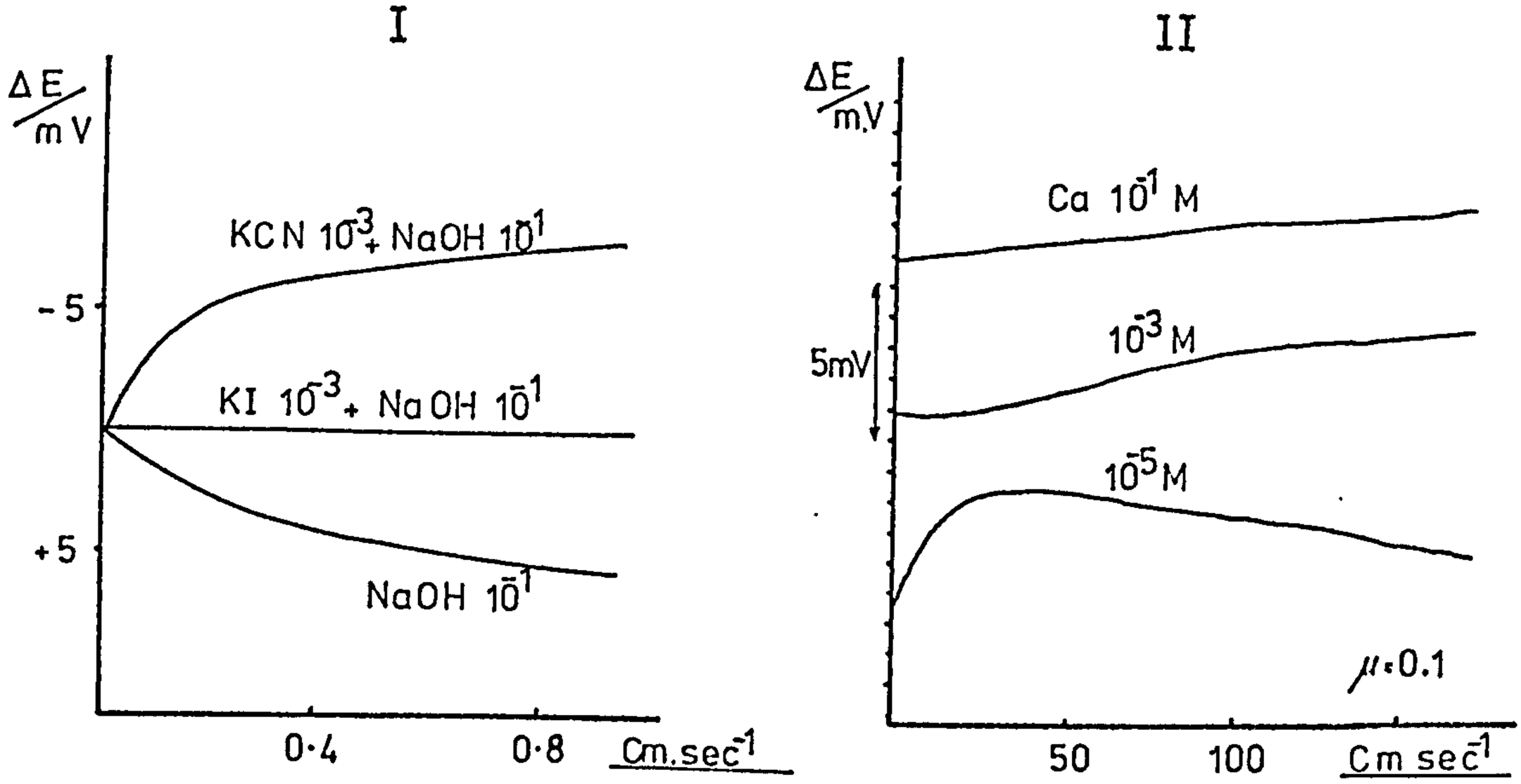
Figure 2.6.1.-I

Differential Titration Cell

2.6.2. Non-Streaming Potential Dependence on Flow Rate

As the equilibrium value of cyanide activity and hence electrode response is established via a diffusion layer at a silver iodide membrane, then a dependence of the potential on flow rate is to be expected. This dependence has been confirmed by evaluating the iodide electrode under flow stream conditions (75). An increase in the flow rate of cyanide solution gives a potential change indicating a higher cyanide activity whereas a similar change with iodide solution produces a very small difference as the diffusion layer is of lesser importance.

Figure 2.6.1.-I/II



Electrode Response to Solution Flow Rate

Although the response is faster, there is no evidence to suggest that, in continuous monitoring, solid state or immobilized membrane electrodes give a more stable response than conventional liquid membranes. A study of an Orion liquid membrane electrode used as a sensor for kinetic measurement of metal ion-ligand combination reactions (75) showed the electrode to give a stable response up to high flow rates (Figure 2.6.2.-II), where deviation was probably due to deformation of the membrane.

In both of these cases the flow rates used in most continuous analysis applications are in plateau regions of the E.M.F./flow dependence, and thus minor fluctuations in the pumping rate (or stirring rate) will not give rise to a large error.

CHAPTER 3

Reference Electrodes

3.1. Internal Reference Electrodes

3.2. "Reference" Electrodes

3.2.1. Reference Electrodes of Constant Potential

3.2.2. Ion Selective Electrodes As Reference Electrodes

Measurements with ion selective electrodes are usually carried out in a cell of the type:
Internal Reference Electrode /a_i/ Membrane/ Sample Solution a_i // Reference Electrode. Thus all membrane potentials are determined with respect to two reference electrodes. In order to ensure that a steady potential can be obtained, the reference electrodes should satisfy several main requirements (76):

i) Ease of preparation from readily available materials,

ii) Rapid attainment of an accurately reproducible potential,

iii) Invariance of potential over long periods of time,

iv) Negligible thermal hysteresis,

v) Low polarizability

Reference electrodes can be divided into two main classes, internal reference electrodes and reference electrode half cells of constant potential.

3.1. Internal Reference Electrodes

The internal reference of an ion selective electrode must be small, rugged, inexpensive and easy to

produce. These requirements are most easily met by the silver, silver chloride electrode. As an electrode of the second kind the half cell is simply a silver wire with a coating of silver chloride, usually formed by electrode-position. In an environment with a constant chloride activity the electrode will give a potential from the reaction:-

$$AgCl(s) \rightleftharpoons Ag^{+} + Cl^{-}$$

$$Ag^{+} + e \rightleftharpoons Ag(s)$$

or $$AgCl(s) + e \rightleftharpoons Ag(s) + Cl^{-}$$

The standard potential in molar hydrochloric acid at 25°C is 0.2224 V which will be incorporated into the value of Eo for the membrane electrode. The silver, silver chloride electrode is used as an internal reference with membranes such as the calcium and potassium liquid and immobilized electrodes. With the pressed pellet silver salt membranes the most frequently used reference system is a silver or silver, silver chloride wire immersed in a silver nitrate solution, facilitating exchange with the silver ions transporting charge in the membrane. An internal reference effect can also be achieved by contacting the silver wire directly to the membrane. Due to the mechanism of reversible ion exchange at the solution-salt interface such an electrode will function as an electrode of the second kind provided that rapid and reversible ion exchange takes place at the metal-salt interface (77).

3.2. "Reference" Electrodes

Reference electrodes are necessary to complete an electrical circuit with a membrane electrode and to

provide a fixed potential against which the E.M.F. of an ion selective electrode can be measured. There are two ways of providing this fixed potential, either by using a reference electrode of constant potential or an ion-selective electrode in a constant environment.

3.2.1. Reference Electrodes of Constant Potential

Reference electrodes of constant potential are anion reversible electrodes of the second kind. The two types most frequently used are the silver, silver chloride electrode, which is also used as an internal reference electrode, and the calomel electrode. The calomel electrode is composed of a layer of calomel paste on mercury with a platinum contact, in a chloride ion environment. Although 3.5 N potassium chloride is sometimes used as the covering solution, saturated, normal and 0.1 normal solutions of potassium chloride are most commonly used. The saturated solution has the advantage that it forms its own salt bridge. The standard potentials of these reference electrodes are given in Table 3.2.1.-I.

Table 3.2.1.-I Standard Potentials of Calomel Reference Electrodes

Covering Solution	Potential (in volt, t = ^{o}C)
0.1 N KCl	+0.336 - 0.00006 (t-25)
1 N "	+0.283 - 0.0002 (t-25)
Satd. "	+0.244 - 0.0007 (t-25)

3.2.2. Ion Selective Electrodes as Reference Electrodes

Although membrane potentials are most frequently measured with respect to a "reference" electrode, they may

also be measured with respect to another selective electrode, provided that this electrode is responsive only towards an electrolyte which is present at a constant concentration. The use of I.S.E.'s as reference electrodes has the added advantage that liquid junction potentials and their associated errors can be avoided, but special differential amplifiers with two high impedance inputs must be used.

SECTION II

DESIGN, PREPARATION, EVALUATION AND APPLICATION

OF ION SELECTIVE ELECTRODES

CHAPTER 4

Commercial Electrode Designs

4.1. Pressed Pellet and P.V.C. Immobilized Electrodes

4.1.1. Electrodes with Filling Solution Internal Contact

4.1.2. Electrodes With Solid Internal Contact

4.1.3. Combined Membrane - Reference Electrodes

4.2. Liquid - Membrane Electrode Design

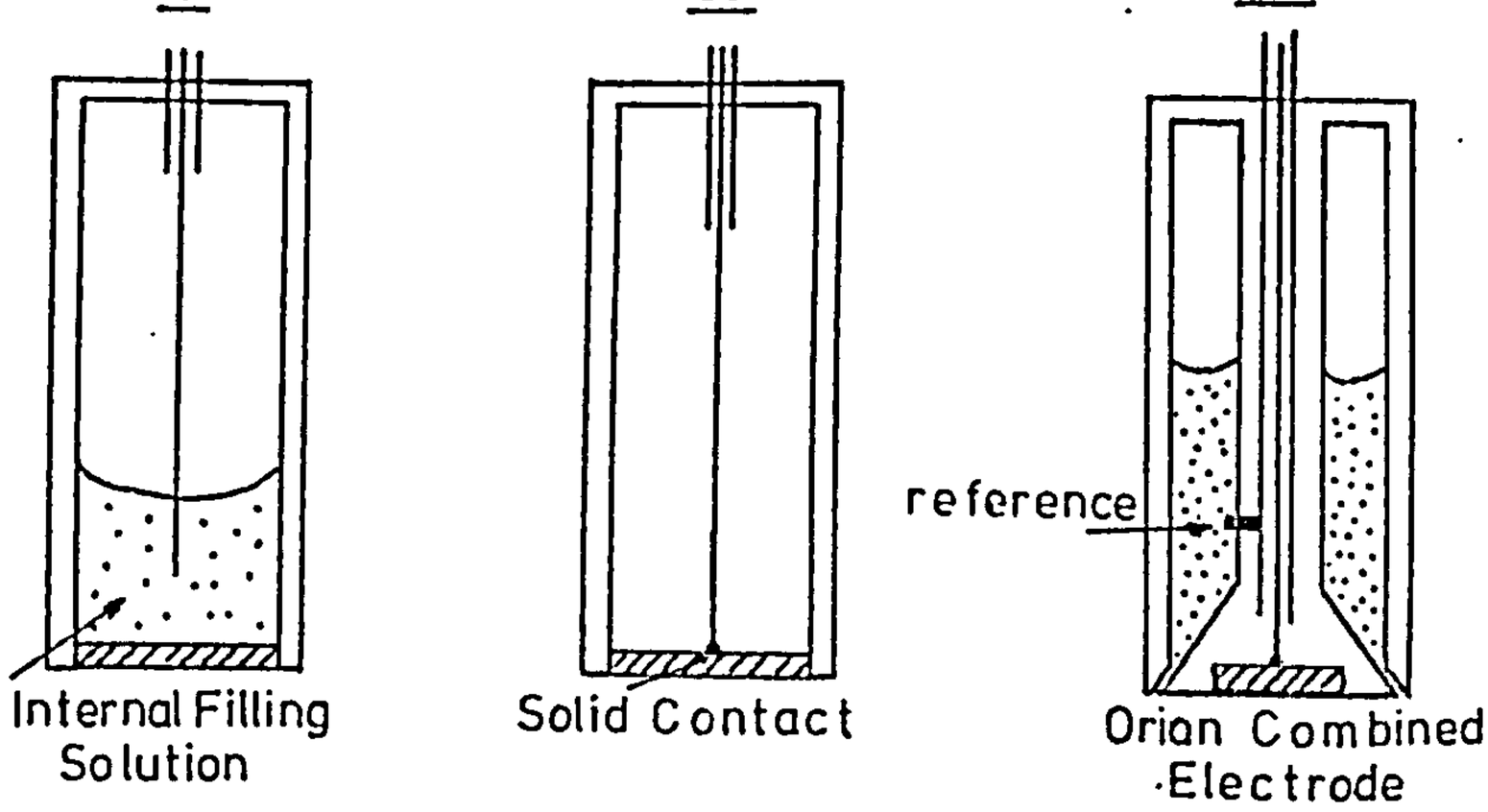
The sensor electrodes can most conveniently be divided into groups of (i) liquid, (ii) immobilized, and (iii) solid-membrane electrodes.

4.1. The Design of Pressed Pellet and P.V.C. Immobilized Membrane Electrodes

4.1.1. Electrodes With Filling Solution Internal Contact

The pressed pellet heterogeneous, homogeneous and P.V.C. immobilized membrane electrodes using an internal filling solution have a similar form. The membrane is stuck with a non-conducting adhesive to a tube of inert material such as glass or epoxy resin in a basic design, shown in Figure 4.1.-I.

Figure 4.1. Commercial Membrane Electrode Designs



4.1.2. Electrodes With Solid Internal Contact

When a liquid filling solution is used and the electrode is inverted, the electrical contact between the membrane and the internal reference electrode may be broken. To avoid this the filling solution is sometimes made into a gel with the addition of a polymer, but as these gels are liable to shrink and cause malfunction of the electrode, much work has been done to make 'dry' or solid contacts direct to the membrane surface. Silver loaded epoxy resins have been found the most effective contact for silver precipitate pressed membranes, Figure 4.1.-II. The electrode designed by Růžička, the Selectrode (43,44) is a solid contact system using the electroactive exchanger as a thin layer rubbed onto the surface of a hydrophobitised graphite support. The solid contact, simply a steel wire screwed into the graphite bar, has been used successfully. Direct contact with P.V.C. immobilized membranes has been achieved in two ways: The simplest contact has been made by holding a graphite rod against a membrane at a constant pressure (42), but as the potential at the internal surface cannot be well defined, the value of Eo is liable to drift. P.V.C. immobilized electrodes have also been made by coating rigid supports such as platinum wire (78) and by coating a silver, silver chloride wire that has previously been coated with a reference electrolyte immobilized in poly (vinyl alcohol) (79).

4.1.3. Combined Membrane Reference Electrodes

The solid state electrodes have been further

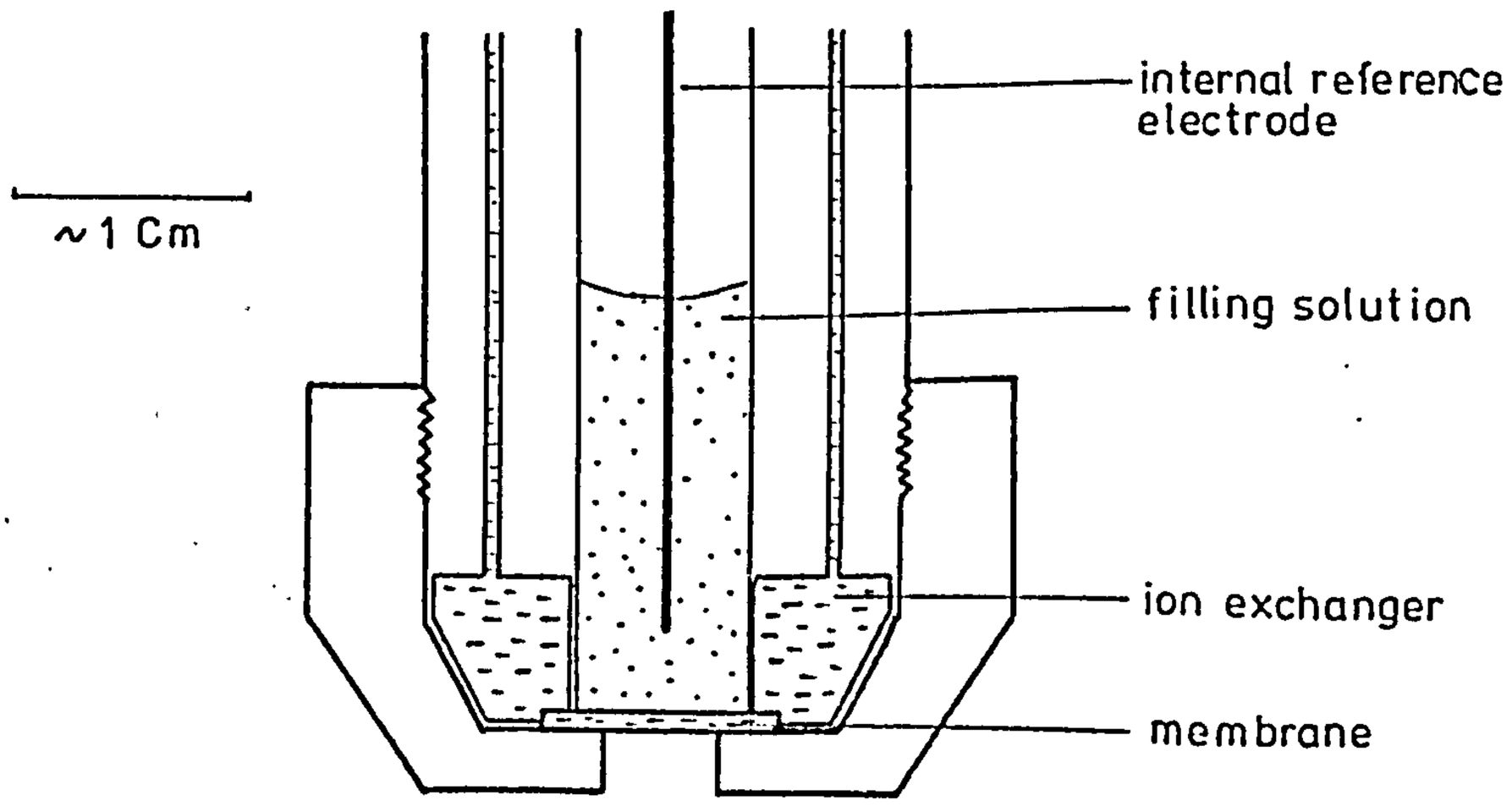
developed by Orion in the production of a combined membrane/ reference electrode. The sensing membrane electrode body is surrounded by a sleeve junction reference electrode (Figure 4.1.-III) allowing the electrode to be used in very small quantities of sample solution, for example the measurement of chloride in sweat.

4.2. Liquid-Membrane Electrode Design

The most common electrode using liquid exchangers is that produced by Orion, with a configuration shown in Figure 4.2.-I. The electrode body is designed to have a reservoir for the exchanger in contact with the porous membrane, so that the exchanger in the membrane is continuously replaced. The internal reference solution is contained in a separate chamber in contact with the internal surface of the membrane. This type of electrode can be used with charged ion exchangers or neutral carrier exchangers dissolved in suitable hydrophobic solvents.

Figure 4.2.-I

Liquid Membrane Electrode Design (Orion)



CHAPTER 5

Sensor Designs Developed In This Study

5.1. Electrodes Designed for Use With Internal Filling Solutions

5.2. Electrodes Designed for Use With Solid Contacts

5.2.1. Pressed Pellet Membrane Electrodes

5.2.2. Immobilized Exchanger Membrane Electrodes

5.3. Combination Membrane-Reference Electrodes

5.1. Electrodes Designed for Use With Internal Filling Solutions

As ion selective electrodes sensitive to several different ions are frequently used, and the membranes of immobilized exchangers have a limited life time, it was thought that an electrode with a replaceable membrane unit would be appropriate. This would also allow easy replacement or "topping up" of the internal filling solution to be done. The electrode design found most useful for this purpose is shown in Figure 5.1.-I.

The pressed pellet membranes were stuck into a "Delrin" tube with 'Araldite' to form a replaceable electrode end cap. This end cap was large enough to contain the reference electrolyte and was threaded to facilitate assembly to a rod of the same material (the electrode 'body') which incorporated a reference electrode and copper screening. The reference electrode was a silver wire spiral with a silver chloride coating formed by making it the anode of an electrical circuit in a 0.1 N solution

of hydrochloric acid. The electrode end caps for the P.V.C. immobilized electrodes were turned down from P.V.C. rod, thus enabling the membranes to be stuck to the caps with P.V.C. dissolved in tetrahydrofuran (T.H.F.) to form an effective seal. The end cap/electrode body design is especially useful if I.S.E.'s responsive to many different ions are needed. One electrode body with a range of end caps can be used with a considerable saving in material costs and manufacturing time.

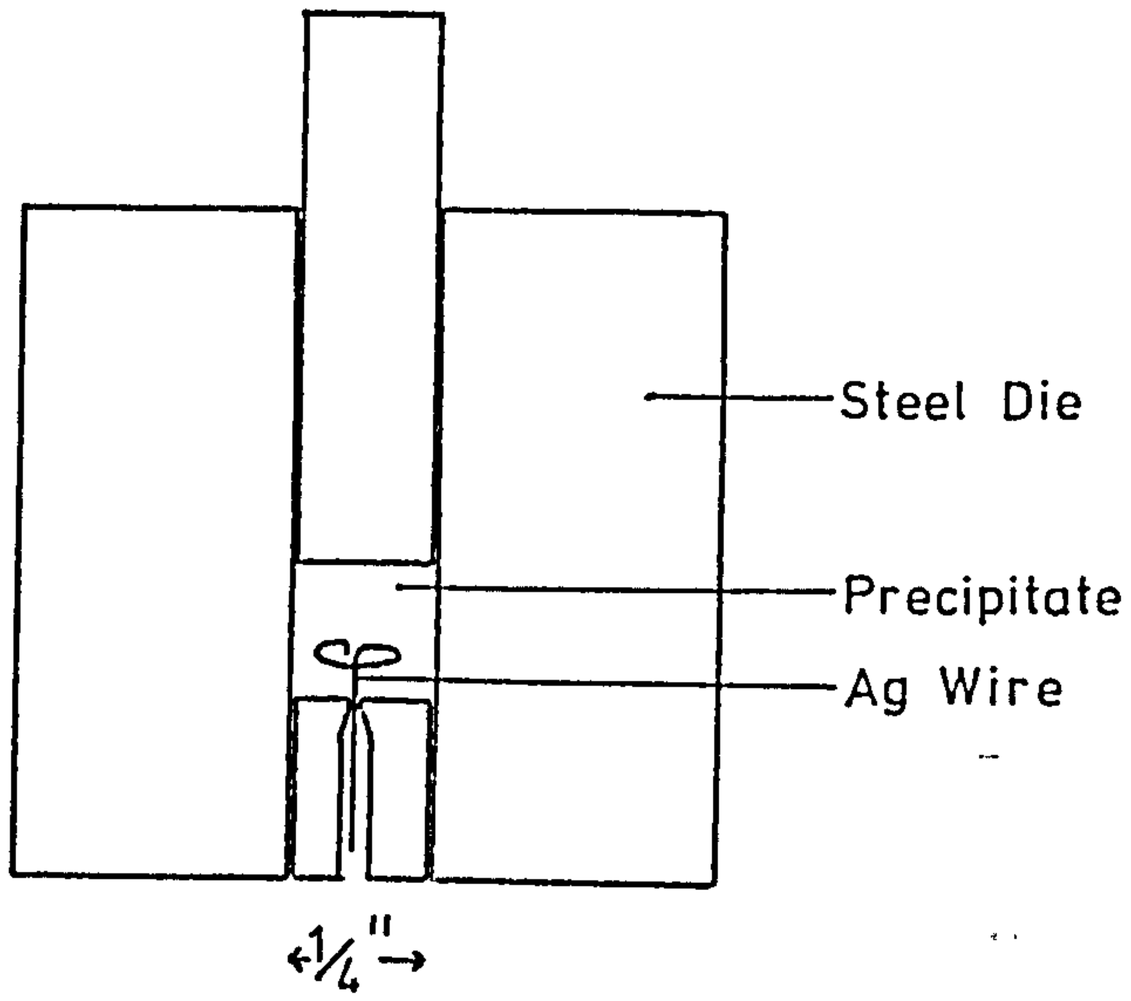
5.2. Electrodes Designed for Use With Solid Contacts

5.2.1. Pressed Pellet Membrane Electrodes

The electrodes designed with a solid contact were primarily intended for use in flow systems, where their use in an inverted position would prevent the accumulation of air bubbles on the membrane surface. Silver epoxy was not an effective contact for the silver iodide membrane and although silver sulphide loaded epoxy was found to work, the best contact system for all silver precipitate based electrodes was found to be a silver wire pressed into the membrane. A stainless steel die was designed with one pellet having a hole drilled through it to accept a spiralled silver wire. The die, with a configuration shown in Figure 5.2.-I was put under sufficient pressure to convert the precipitate to a single crystal (3 tons load) in an infra red pellet press. When the membrane was removed from the die, it was stuck into a 'Delrin' electrode end cap which was modified for use with a 'solid state' electrode body (Figure 5.2.-II).

Figure 5.1.-I Electrode Design Incorporating A Filling Solution

Figure 5.2.-I



Die for Solid Contact Electrodes

The contact system was found to work effectively with all of the membranes used (AgCl, AgBr, AgI, Ag_2S) without changing the electrode performance other than the value of E_o.

5.2.2. Immobilized Exchanger Membrane Electrodes

To enable the P.V.C. immobilized membranes to be used inverted, the electrode body rather than the electrode end cap was modified. A carbon (spectroscopic grade graphite) rod was stuck into the body so that it could be held in contact with the membrane by the screw threads (Figure 5.2.-III). Although this contact enabled

Figure 5.2.-II Precipitate Based Solid State Membrane Electrodes

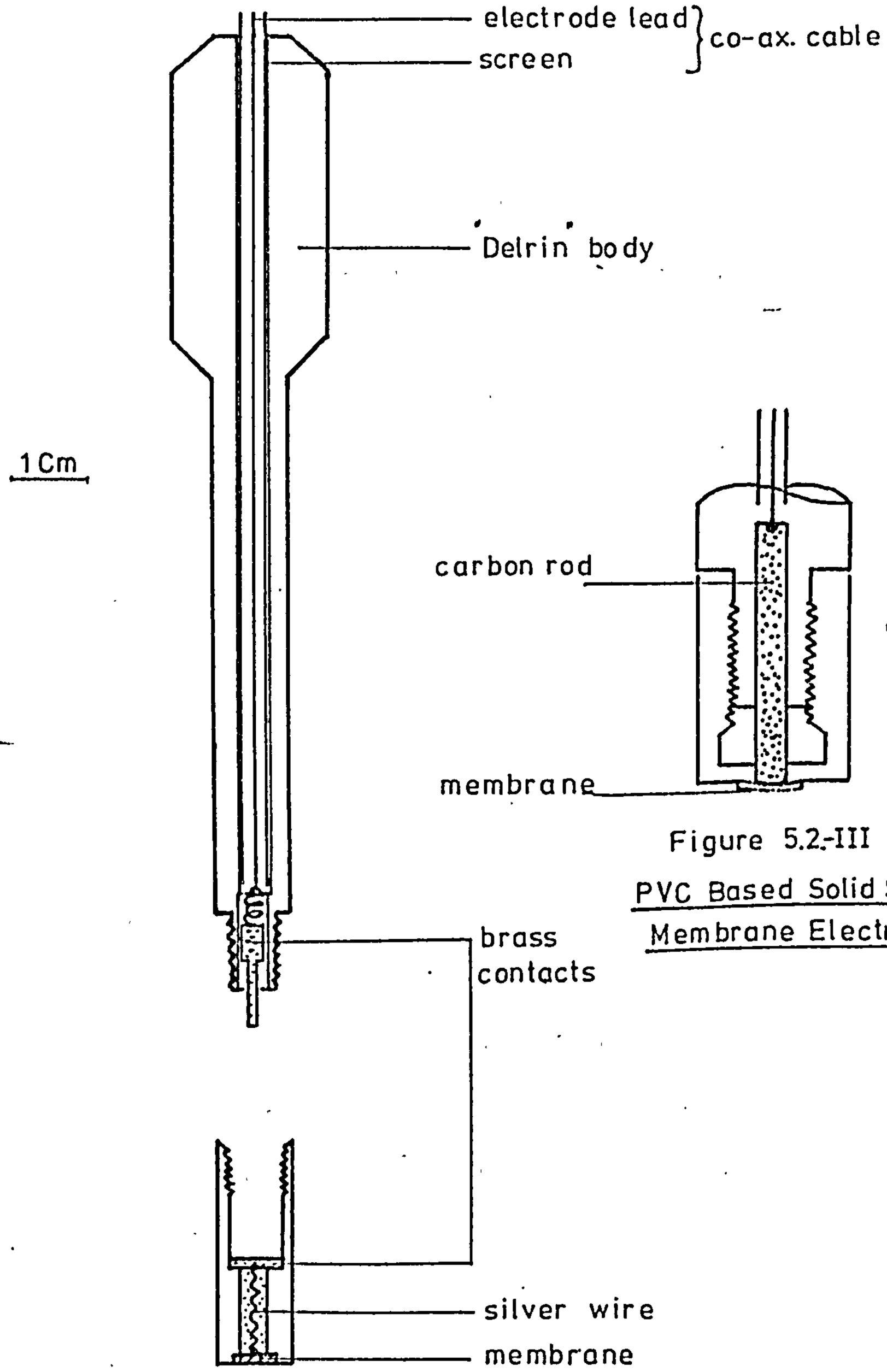
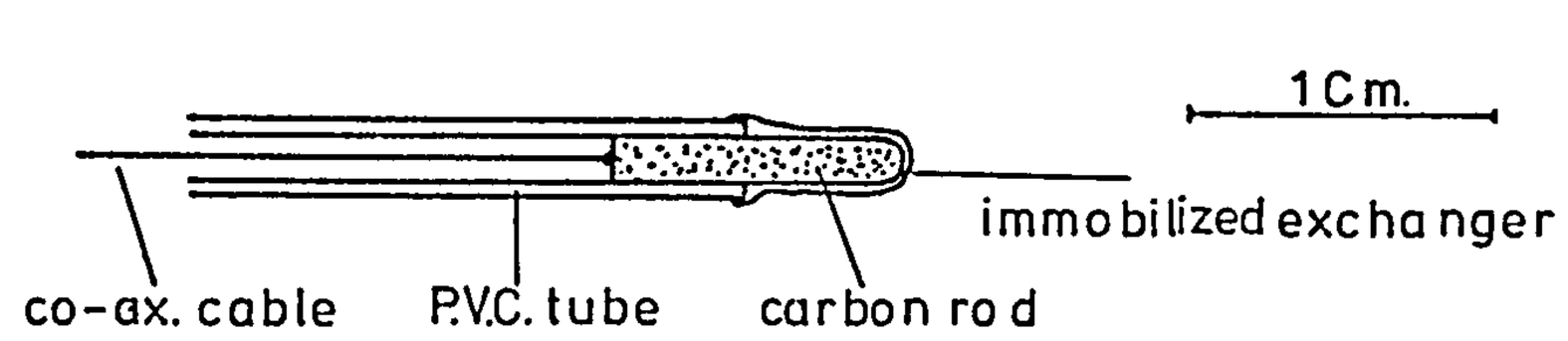


Figure 5.2.-III
PVC Based Solid State Membrane Electrodes

short term work to be done, the value of E_o was so variable that the electrode needed frequent recalibration. The response slope was reproducible however and the electrode could be used for applications such as standard addition potentiometry and titration end point monitors.

As an attempt at production of very cheap disposable electrodes, P.V.C. coated graphite electrodes were made (Figure 5.2.-IV). The graphite rod was simply dipped into a P.V.C. membrane mixture and allowed to dry. Both lead and calcium electrodes were made successfully using this system.

Figure 5.2.-IV Coated Graphite Electrodes



5.3. Combination Membrane/reference Electrodes

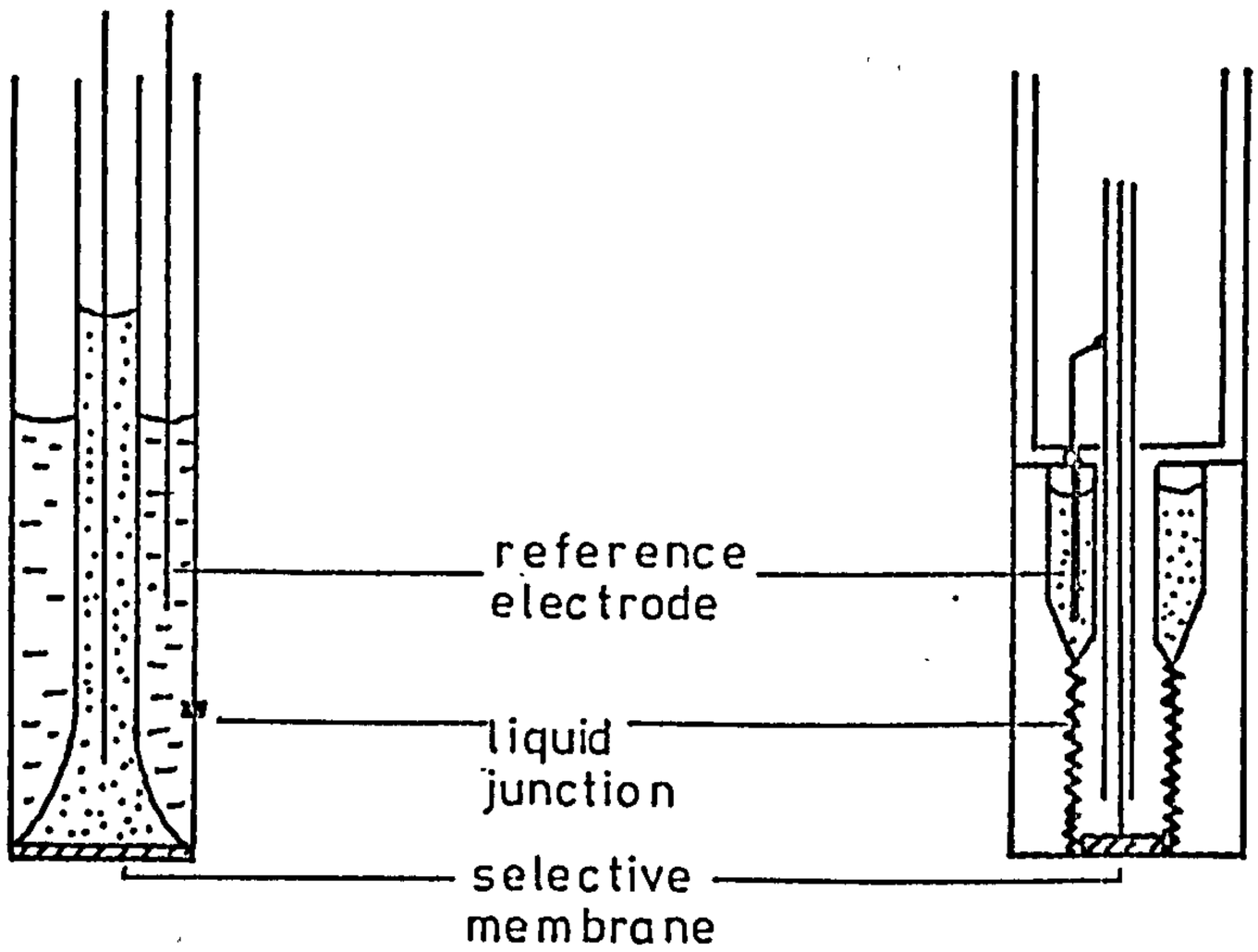
Early combination electrodes were made by modification of broken glass electrodes. The pH sensitive glass was removed gently with fine emery paper and a flat surface was prepared. The internal reservoir was filled with reference electrolyte and the membrane (e.g. a single crystal of lanthanum fluoride) was stuck in place with 'Araldite'. This method was successful in the preparation of combined electrodes sensitive to both fluoride and iodide although both were obviously fragile (Figure 5.3.-I). In order to reduce the sample volume

needed for analysis, the distance between the reference junction and sensing membrane must be reduced. This was achieved by bringing the junction and membrane into the same plane, using screw threads and "tight fits" as liquid junctions (Figures 5.3.-II, III).

When a combined chloride/reference electrode was required, it was necessary to incorporate a salt bridge to prevent the leakage of reference electrolyte into the sample solution and an electrode was developed using "tight fits" as junctions in the design, shown in Figure 5.3.-IV. The use of P.V.C. or 'Delrin' as the material in these designs makes the electrodes compact and virtually unbreakable.

Figure 5.3.-I

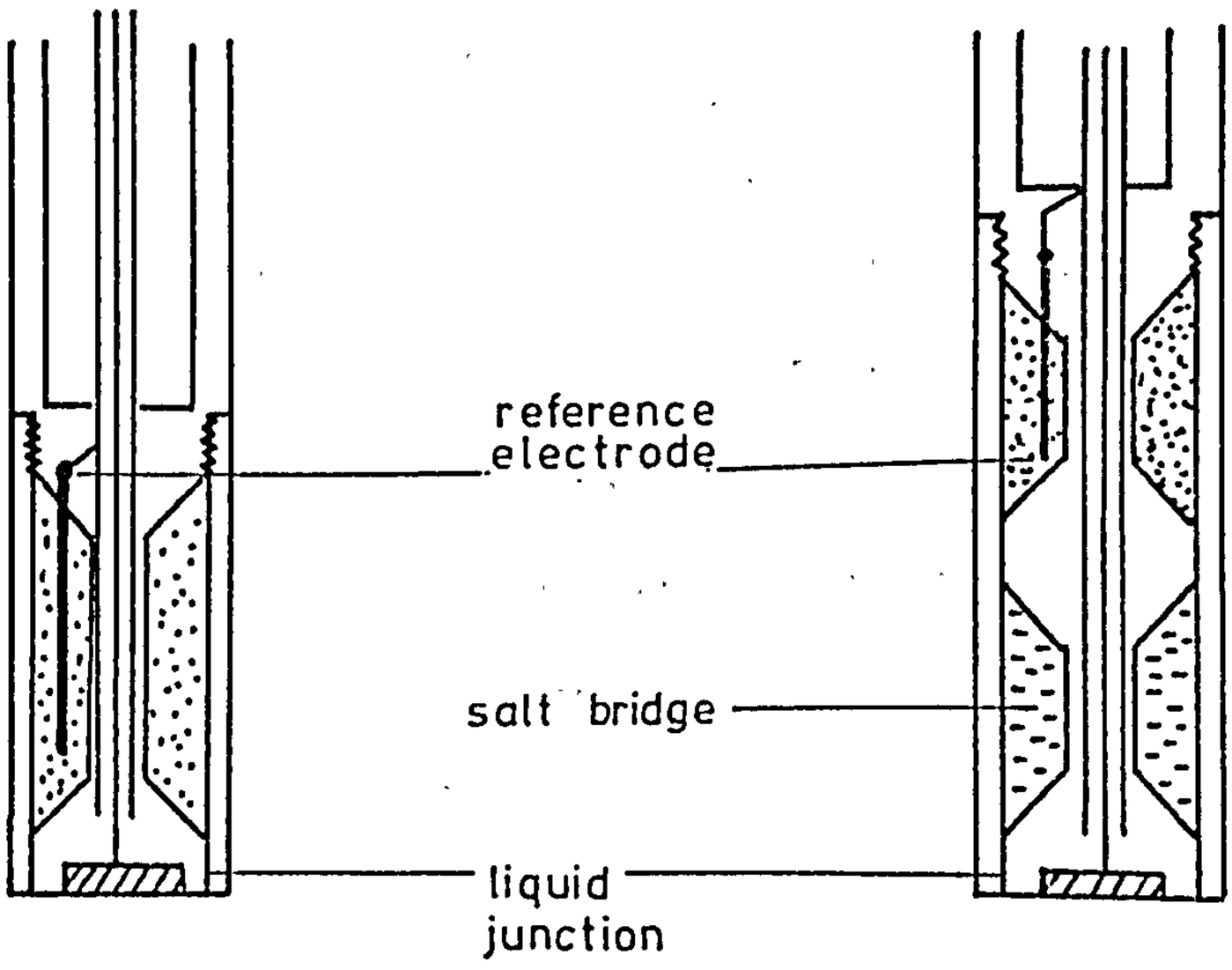
Figure 5.3.-II



Combined Membrane and Reference Electrode Designs

Figure 5.3.-III

Figure 5.3.-IV



CHAPTER 6

Preparation Of Membranes For Ion Selective Electrodes

6.1. Pressed Pellet Membranes

6.1.1. Anion Sensitive Membranes

6.1.2. Cation Sensitive Membranes

6.2. P.V.C. Immobilized Electrodes

The electrodes prepared for use in this study can be divided into two classes, those including pressed pellet membranes and those including poly (vinyl chloride) immobilized membranes.

6.1. Pressed Pellet Membranes

The pressed pellet membranes prepared were all based on silver salts, usually of similar composition as the electrodes described by Orion (37,80). Both anion and cation sensitive electrodes contained silver sulphide to decrease membrane resistance and the effects of sunlight.

6.1.1. Anion Sensitive Membranes

The Chloride, Bromide and Iodide membranes were all produced using a similar method. Molar solutions were made up of the potassium salt of the halide, and sodium sulphide. The solutions were mixed in a ratio 9(halide) : 1(sulphide) and the resulting solution was added slowly to molar silver nitrate with vigorous stirring. The granular precipitate obtained was filtered off, washed with distilled water until free of excess silver, and then alcohol, before drying in an oven at 80-100^{o}C. It was found that electrodes prepared by adding the silver nitrate

solution to the mixed salt solution tended to give non Nernstian slopes and frequently a reduced response range.

The sulphide responsive membrane precipitate was prepared by passing hydrogen sulphide through silver nitrate solution, or by adding sodium sulphide.

The dry precipitates (0.2-0.3g) were placed in a KBr pellet press 1/4" in diameter and subjected to a load of three tons for five minutes under vacuum. The sulphate selective electrode was prepared according to the directions given by Rechnitz et. al. (81).

6.1.2. Cation Sensitive Membranes

When cation sensitive membranes were prepared, the procedure described by Czaban and Rechnitz (38) was followed. Appropriate volumes of molar silver nitrate and metal nitrate were mixed and added to excess sodium sulphide. The precipitate was washed with hot water and acetone then filtered and dried. In the case of the lead membrane, the precipitate was first boiled, filtered and washed with dilute nitric acid, then washed with water and mixed with carbon disulphide to prevent the formation of lead sulphate. Membranes selective towards lead and copper were produced in this way.

6.2. P.V.C. Immobilized Electrodes

An investigation of the optimum composition of poly (vinyl chloride) matrix membranes, used for calcium selective electrodes, has been carried out by Griffiths, Moody and Thomas (82). 28.8 per cent w/w of poly (vinyl chloride) and 71.2 per cent w/w of ion exchanger were found

to give the best calcium electrodes. Based on these results P.V.C. immobilized membranes were prepared in the following way:

Poly (vinyl chloride) (0.09 gms) was mixed with the ion exchanger solution (0.3g) which was either a commercial or prepared liquid exchanger. Sufficient dry tetrahydrofuran (T.H.F.) to dissolve the P.V.C. was added and the mixture was poured into a glass ring (2.5cm I.D.) on a glass plate. The ring was covered by weighted filter papers and left for 48 hours. The membrane plug prepared in this way was rubbery and resilient, being about 0.5mm thick. Electrode membranes were cut from this plug using a No. 2 cork borer and were mounted onto tubing with P.V.C. dissolved in T.H.F.

CHAPTER 7

Evaluation of the Membrane Electrode

7.1. Sensitivity

7.2. Selectivity

7.2.1. Selectivity by Separate Solution Methods (I)

7.2.2. Selectivity by Mixed Solution Methods (II)

The value of an electrode for analysis can be ascertained from the sensitivity and selectivity of the membrane. The sensitivity of the membrane can be defined as whether or not the membrane will respond to the ion under investigation over the concentration range necessary, and the selectivity of the membrane as whether or not the presence of other electrolytes in the sample will prevent accurate analysis of the unknown. Thus if an ion selective electrode is to be used for analysis its sensitivity and selectivity must be known.

7.1. Sensitivity

The sensitivity of an electrode to a particular ion is simply determined by introduction of the membrane, with a suitable reference electrode, into several different solutions of the ion concerned. As the electrode theoretically obeys the Nernst equation (ignoring inter-ferences):

$$E = E_o + \frac{RT}{nF} \ln a$$

the potential of the membrane will vary linearly with the logarithm of the ion activity, and a graph of E (in mv) against $\log_{10}$ ai will be linear with a slope of 2.303 RT/nF

(59.16 for a univalent ion (n = +1) at 25°C), over a wide range of ion activities. Concentration calibrations are not necessarily straight lines and may have a slope different from that of the activity curves. This is due to the influence of ionic strength on the activity. The relationship between the activity and the concentration is given by the equation 7.1.- 1 .

$$a_i = \gamma_i \; C_i \qquad 7.1.-1$$

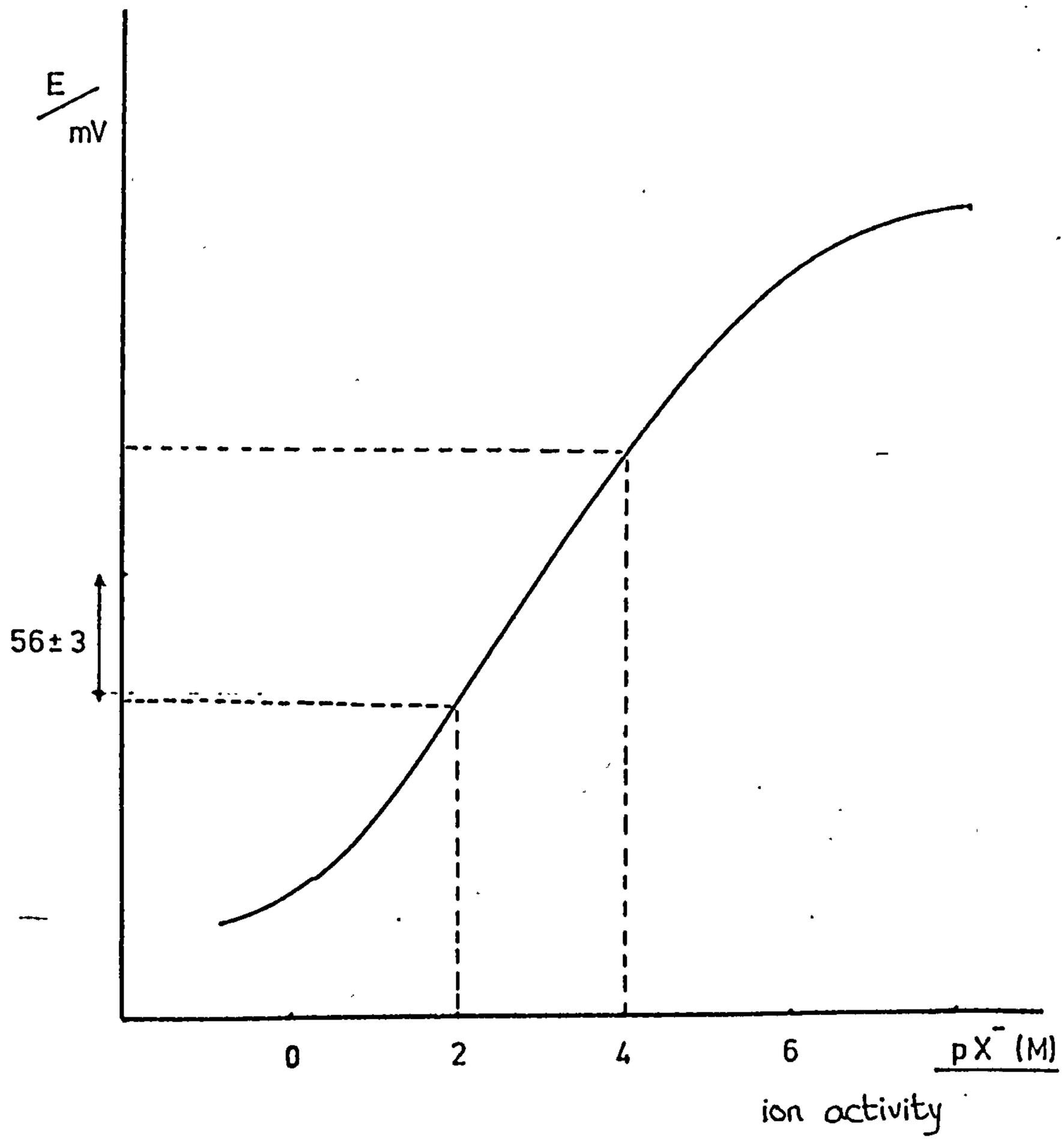
where a_i = activity of the ion, γ_i = activity coefficient of the ion and C_i = the concentration of the ion, in molarity. The activity coefficient depends on the total ionic strength of the solution (μ) which is defined as "half the total sum of the concentration of each and every ion in the solution (C) multiplied by the corresponding square of its charge,

$$\mu = 0.5 \sum Z^2 C \qquad 7.1.-2$$

Values of γi are tabulated by Keiland (83) and were used throughout this work.

If an ionic strength buffer such as T.I.S.A.B. is added to the analyte and standard solutions, to keep the activity coefficient of the ion constant, activity may be replaced in the Nernst equation, and the electrode calibrated directly, in terms of concentration. In both cases the graph will deviate from linearity at low concentrations, in the region of the detection limit, and at high concentrations, where the ionic activity cannot be well defined, as is shown by a typical calibration graph in Figure 7.1.-I.

Figure 7.1.-I Typical Electrode Calibration Graph



7.2. Selectivity

To obtain the full use of an ion selective electrode it is necessary to know the selectivity toward ions in the analyte in order to evaluate their interference.

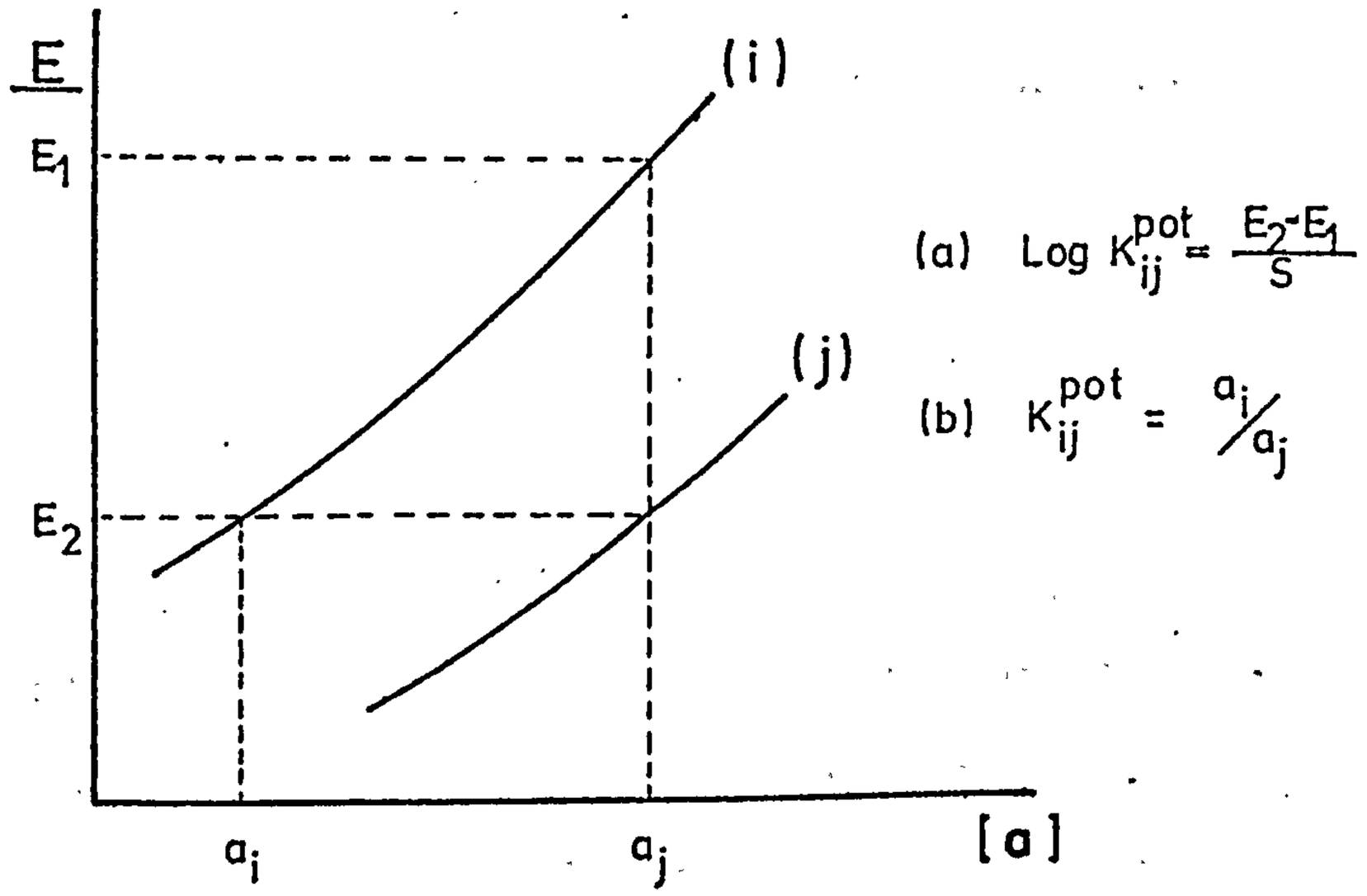
The selectivity has already been defined by K^{pot}_{ij} (eqn.2.2.2.-2), where:

$$E = E_o + \frac{RT}{nF} \ln a_i^{n+} + K^{pot}_{ij} (a_j{}^{z+})^{n/z} \qquad 7.2.-0$$

For all values of $K^{pot}_{ij} < 1$ the electrode favours species (i) over species(j). Various methods for evaluating the selectivity ratios, based on potential measurements in both separate and mixed solutions have been discussed (84) and the techniques used in this study are described below:

7.2.1. Selectivity by Separate Solution Methods (I)

Figure 7.2.-I Separate Solution Methods of Selectivity Evaluation



A calibration graph for both the primary cation and the interfering ion is made, (Figure 7.2.-I) and from this selectivities can be evaluated in two ways:

(a) The response E_1 to a primary cation is:

$$E_1 = Eo + S \log a_i \quad (S \quad \frac{RT}{nF} \times 2.303) \qquad 7.2.-1$$

and the response to the interfering ion (j) in the absence of primary ion (i) is given by equation (7.2.-0) with $a_i = 0$.

$$E_2 = E_o + S \log K^{pot}_{ij} a_j \qquad 7.2.-2$$

Subtracting equation (7.2.1) from (7.2.2.) gives:

when $a_j = a_i$

$$\frac{E_2 - E_1}{S} = \log K^{pot}_{ij} \qquad 7.2.-3$$

The values E_2 and E_1 are easily obtained from the calibration plots at any activities $a_i = a_j$

(b) When $E_1 = E_2$ equation (7.2.1.) and (7.2.2.) can be combined to give $\log a_i = \log K^{pot}_{ij} a_j$ 7.2.-4

i.e. $a_i = K^{pot}_{ij} a_j$

With different charges on primary ion (n^+) and interferent ion (Z^+) the equation is modified to

$$a_i^{n+} = K^{pot}_{ij} (a_j)^{z/n} \qquad 7.2.-5$$

Neither of these two methods is realistic since separate solutions are used (85).

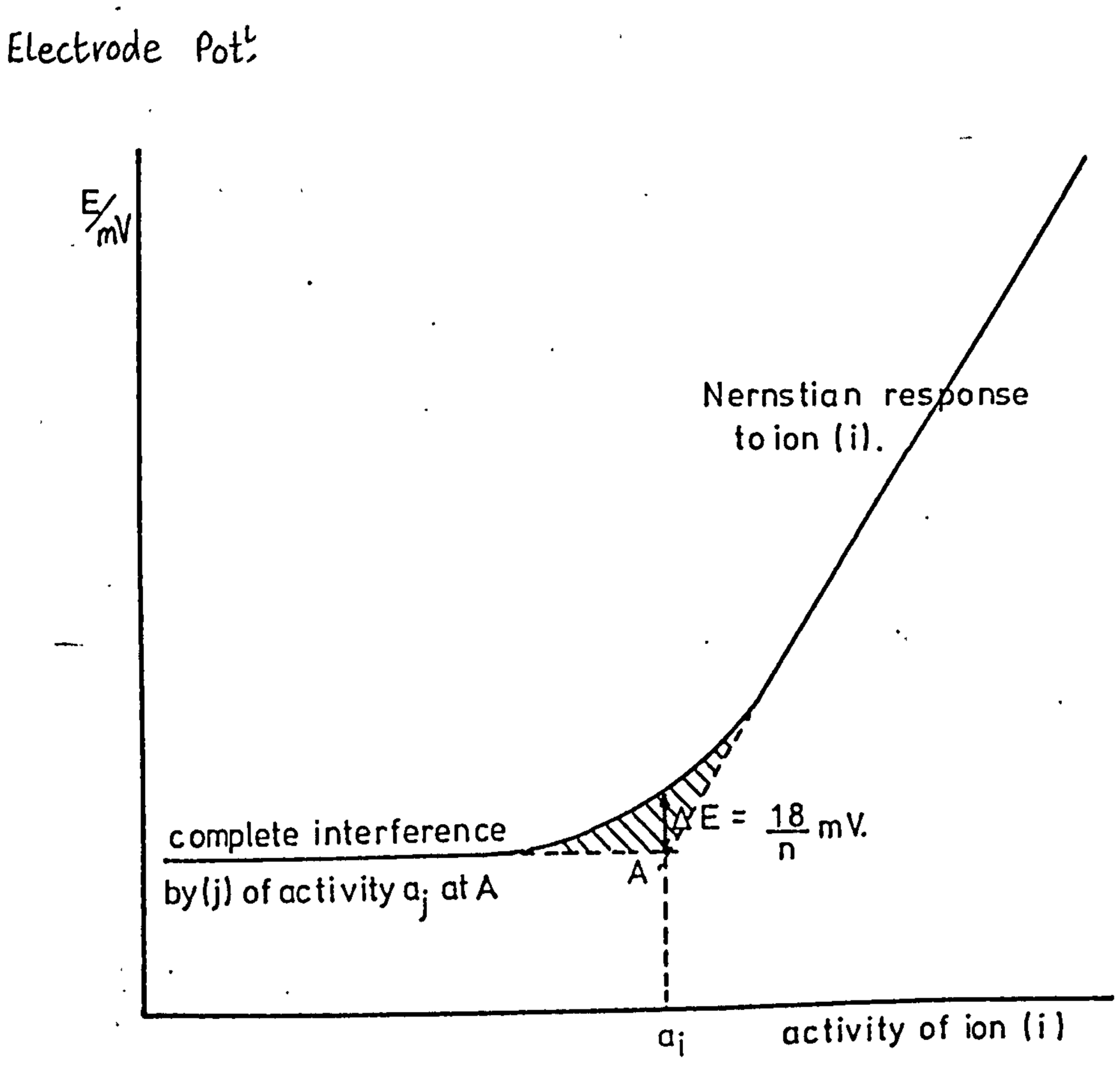
7.2.2. Selectivity by Mixed Solution Methods (II)

a) A common procedure is to measure potentials in solutions containing a fixed initial concentration of interferent (j^{2+}) while varying the activity of the primary ion (i^{2+}) (Figure 7.2.-II).

As the activity of the primary ion is increased the potential gradually breaks away from the interference level (shaded zone) until a Nernstian response to primary ion is achieved. The intercept of the extrapolated Nernstian

response with the interference plateau defines a particular activity of the primary ion a_i. The selectivity is then calculated from $a_i = K^{pot}_{ij} a_j$ where a_j is the background interference level.

Figure 7.2.-II Illustration of Selectivity Constant (K^{pot}_{ij}) Evaluation Using Fixed Initial Concentration of Interferent



$$K^{pot}_{ij} = a_i / a_j$$

When the plateau region is illdefined the required value (a_i) can be found by taking the point on the curve where ΔE between the curve and extrapolated Nernstian line is $^{18}/_{n}$ mV. The activity at this point will be the required value a_i^{n+} (86).

b) As an alternative the interferent activity can be varied using a constant level of primary ion. The results are treated in a similar fashion to those in method IIa.

c) Srinivasan and Rechnitz (87) use a graphical evaluation of two complex equations for high and low selectivities respectively.

The potential E_1 is measured in the pure solution of the primary univalent anion (i^-) at activity a_i and $E_1 = E_o - S \log a_i$ $\quad (S = \frac{2.303\ RT}{F})$ 7.2.-6

Known quantities of univalent interferent j^- are successively introduced so as to give a series of potential values for the mixed solutions. The potential, E_2, in any one of the series is related to the new primary activity a_i^1 and interferent activity a_j^1 by :-

$$E_2 = Eo - S \log (a_i^1 + K_{ij}^{pot}\, a_j^1) \qquad 7.2.-7$$

Combining equations (7.2.-6) and (7.2.-7)

$$\exp\left(\frac{E_1 - E_2}{S}\right) a_i - a_i^1 = K_{ij}^{pot}\, a_j^1 \qquad 7.2.-8$$

Plots of the left hand side of equation (7.2.-8) vs. interferent activity (a_j) gives a straight line with slope K_{ij}^{pot}.

Where K_{ij} is small the change in potential caused by an addition of interferent will be small, so the primary ion is added to a high 'constant' level of interferent ion. The selectivity coefficient is given by a graphical analysis of the equation

$$a_i^1 - \left[\exp\left\{\frac{E_1 - E_2}{S}\right\}\right] a_i = K^{pot}_{ij}\left[\left(\exp\left\{\frac{E_1 - E_2}{S}\right\}\right)a_j - a_j^1\right] \qquad 7.2.-9$$

d) It is suggested that the standard additions method of concentration analysis may be used to find the selectivity coefficient. Successive additions of primary ion (i) are made to a known concentration of interferent ion, (C_j), and the resulting potentials are analysed using the computer program ADDFIT* (88) to give an assumed concentration of primary ion ($'C_i$). At low concentrations the activity coefficient approaches unity and $C_i \longrightarrow a_i$, then $K^{pot}_{ij} = {}'C_i/C_j$. At higher concentrations, or for more accurate work, the activity coefficient of the ions will have to be taken into account if they differ to any appreciable amount, $K^{pot}_{ij} = \dfrac{C_i \gamma_i}{C_j \gamma_j}$

All of the selectivity methods described give selectivity coefficients that are activity dependent. Buck has proposed that the selectivity coefficients depend on the activity ratio of interferent and primary ion (89). He indicated two types of behaviour of the 'apparent selectivity coefficient' (K^{pot}_{ij} (app)) as a function of ion activity:-

* see appendix

i) K^{pot}_{ij} (app) is a function only of the activity ratio a_j/a_i, and a plot of K^{pot}_{ij} (app) *vs.* a_j/a_i will give all the information required; this is valid for metal ion interference on a glass pH electrode.

ii) K^{pot}_{ij} (app) is a function of the ratio a_j/a_i and the individual ionic activities a_i, a_j. In this case a family of curves would be necessary to specify the response, e.g. for the lithium error in a Beckman sodium glass electrode.

Therefore, in order to recognize the full value of the term K^{pot}_{ij}, the activity of the primary ion or interferent ion at which it was calculated, and the method by which it was found should be stated.

CHAPTER 8

Analytical Techniques Using Ion Selective Electrodes In Non - Automated Systems

8.1. Direct Potentiometric Methods of Analysis

8.1.1. Direct Potentiometry

8.1.2. Direct Potentiometry with Precalibration

8.1.3. Bracketing Standards

8.1.4. Null Point Potentiometry

8.1.5. Standard Addition Techniques

8.2. Indirect Analysis by Titration

8.3. Differential Potentiometry

Most analytical techniques employing ion-selective electrodes can be divided into two main groups:-

a) Direct potentiometric measurements based on evaluation of the Nernstian logarithmic relationship between cell E.M.F. and ion activity, and,

b) Indirect titrimetric procedures.

8.1. Direct Potentiometric Methods of Analysis

8.1.1. Direct Potentiometry

The potential generated between the reference and indicating electrodes (E_{cell}) is governed by the equation:-

$$E_{cell} = E^{o}_{ind} - E_{Ref} - E_J + \frac{RT}{nF} \ln \left[a_i^{(n+)} + K^{pot}_{ij} (A_j^{Z+})^{n/z} \ldots\ldots \right]$$

where E_{Ref} is the reference electrode standard potential and E_J is the liquid junction potential.

It is not usually possible, however, to use the measurement of E_{cell} to give the activity of the species

i (a_i), as the values of E_o (which includes the internal reference electrode potential) and E_j are unknown and frequently variable. Furthermore, if the concentration of the ion is required, an assumed value must be taken for the activity coefficient.

8.1.2. Direct Potentiometry with Precalibration

The most frequently used technique of analysis avoids absolute measurement of potential by calibrating the electrode in solutions of known ionic activity. The potential obtained in the analyte solution is then measured and the activity obtained by interpolation from the calibration graph. Variations in ionic strength can be avoided by adding a concentrated electrolyte, such as T.I.S.A.B., to both calibrating and unknown solutions. The limitations in accuracy of direct potentiometry resulting from the logarithmic relationship between electrode potential and activity are widely appreciated. An error in activity of 4% is found with a 1 mV change in potential for a univalent electrode at 25°C. In addition to this most electrodes show at least some drift in the magnitude of the constant term (Eo) of the Nernst equation. A computer program called 'Orion' (90) has been produced to minimize the errors associated with direct potentiometry with precalibration while retaining its simplicity and speed.

8.1.3. Bracketing Standards

This technique is identical to precalibration except that the unknown activity is related to two or more

standard solutions which bracket the unknown. The method assumes a linear relationship between cell potential and log. of ion activity, but does avoid the unknown variation of Eo.

8.1.4. Null-Point Potentiometry

In principle, null-point potentiometry is a simple concentration-cell technique that compares the analyte solution with a solution of known composition, (91). The usual procedure involves adjusting the composition of one of the half-cell solutions to match the other, which is indicated by a zero-cell potential between two identical electrodes specific for the ion of interest separated by a liquid junction. This method can achieve high accuracy but only with electrodes of the same response.

8.1.5. Standard Addition Techniques

With discrete samples a high degree of accuracy can be obtained using standard addition methods of analysis (92,93). One or more aliquots of a standard concentration of the species of interest are added to the unknown, ionically buffered sample solution. If the activity coefficient and liquid junction potential are assumed constant, the cell E.M.F. can be expressed by:-

$$E = E^{o} + S \log \left[\frac{CV + \sum_{i=1}^{j} C_i V_i}{V + \sum_{i=1}^{j} V_i} \right] + \left[\begin{array}{l} S \log \gamma E \\ \text{Assumed constant} \end{array} \right] \qquad 8.1.-2$$

where V is a known volume of the test solution of unknown concentration C; C_i is the concentration of standard which is added in aliquots V_i ml. S and E^{o} are respectively the

response slope and standard potential of the electrode system. If the slope is known, only a single addition is necessary to determine C, which is given by

$$C = Cs \times \frac{Vs}{V + Vs} \cdot \left[10^{\Delta E/S} - \left(\frac{V}{V + Vs} \right) \right]^{-1} \qquad 8.1.-3$$

Where V is the volume of the test solution, Vs and Cs are the volume and concentration of the standard added. ΔE is the change in E.M.F. caused by the addition and S is the slope. Unknown concentrations can be computed using ΔE values published by Orion (94).

Multiple addition can be used without prior knowledge of S and E^o however, equation 8.1.-3 can be rearranged to:-

$$(V + Vs)\ 10^{E/S} = (10^{(E^o + E_J)/S}) \cdot \gamma (C\,V + Cs\,Vs) \qquad 8.1.-4$$

Again assuming a constant E_J and γ, a straight line is formed by plotting $(V + Vs)10^{E/S}$ vs. Vs. This straight line intercepts the volume axis at a point V_E, such that : $C\,V = -Cs\,V_E$. This is the basis of the Gran's plot standard addition method (95,96). The graphical procedure tends to eliminate random errors, but the calculation of $10^{E/S}$ is tedious and places a large emphasis on the electrode slope remaining constant throughout the experiment.

A computer program , ADDFIT, by Brand and Rechnitz (88) has been devised to evaluate the concentration, S and E^o using a non-linear, least-squares curve fitting analysis for multiple additions. The main advantages of this technique are summarized below:

1. Although the electrode responds to activity the result is in terms of concentration.

2. No electrode precalibration, or calibration graphs are necessary.
3. It is a fast and simple technique in operation.
4. Only one standard solution is used.
5. It is the only available method of using an I.S.E. to determine concentration of an ion in the presence of large amounts of complexing ion.
6. A similar method can be used based on the subtraction of ion, e.g. by adding a complexing or precipitating agent.

8.2. Indirect Analysis by Titration

In contrast to direct potentiometry, potentiometric titrations generally offer accuracy and precision at the cost of increased time and difficulty. They also allow the determination of species for which no electrodes are available, using a selective electrode responsive to the titrant. In a typical titration the electrode potential is plotted against the amount of either complexing, precipitating, redox or neutralising agent added, and the equivalence point is measured from the curve. The high accuracy of the titration, primarily limited by the standardization and volume measurement of the titrant and by the sharpness of the potential 'break' near the endpoint, is achieved because many of the unknown variables, (Ej, E^{o}_{ind} and E_{ref}) do not play a large part in the position of the equivalence point. In addition potentiometric titrations are feasible in non-aqueous media where direct potentiometric data are difficult to interpret.

The critical problem in potentiometric titrations is obviously the location of the endpoint, (97,98). The potential at the equivalence point can be found by calculation or by using a standard, but for most practical purposes the point of inflection on the curve is located and utilized. If ambiguities exist due to an asymetric titration curve, the inflection point can be located by plotting either the first or second derivative of the curve as a function of the volume of titrant added. When the effects of dilution are included, it has been shown (99) that the inflection point of the titration curve and the theoretical equivalence point do not coincide. The titration error is small however, ($<1\%$), unless a high level of interferent ion is present. A general treatment has been presented which describes the effect of electrode selectivity, interfering ion concentration, sample ion concentration, titration-reaction equilibrium constant and dilution on potentiometric titrations using ion selective electrodes as indicators (100).

8.3. Differential Potentiometry

Differential potentiometry is a technique devised to eliminate liquid junctions. The potential of an ion selective electrode is measured,not against a reference electrode,but with respect to another selective electrode which, if a corresponding constant ionic activity is present in solution, will give a constant potential. The 'differential' approach can be used in many applications including direct potentiometry with precalibration and null point potentiometry, but it is most usefully applied in potentiometric titrations, where two indicating electrodes can have a large voltage difference at the endpoint.

CHAPTER 9

Instrumentation

9.1. High Impedance Voltmeter

9.2. Differential Amplifier

9.3. Antilog Amplifier

9.4. Recorder

9.1. High Impedance Voltmeter

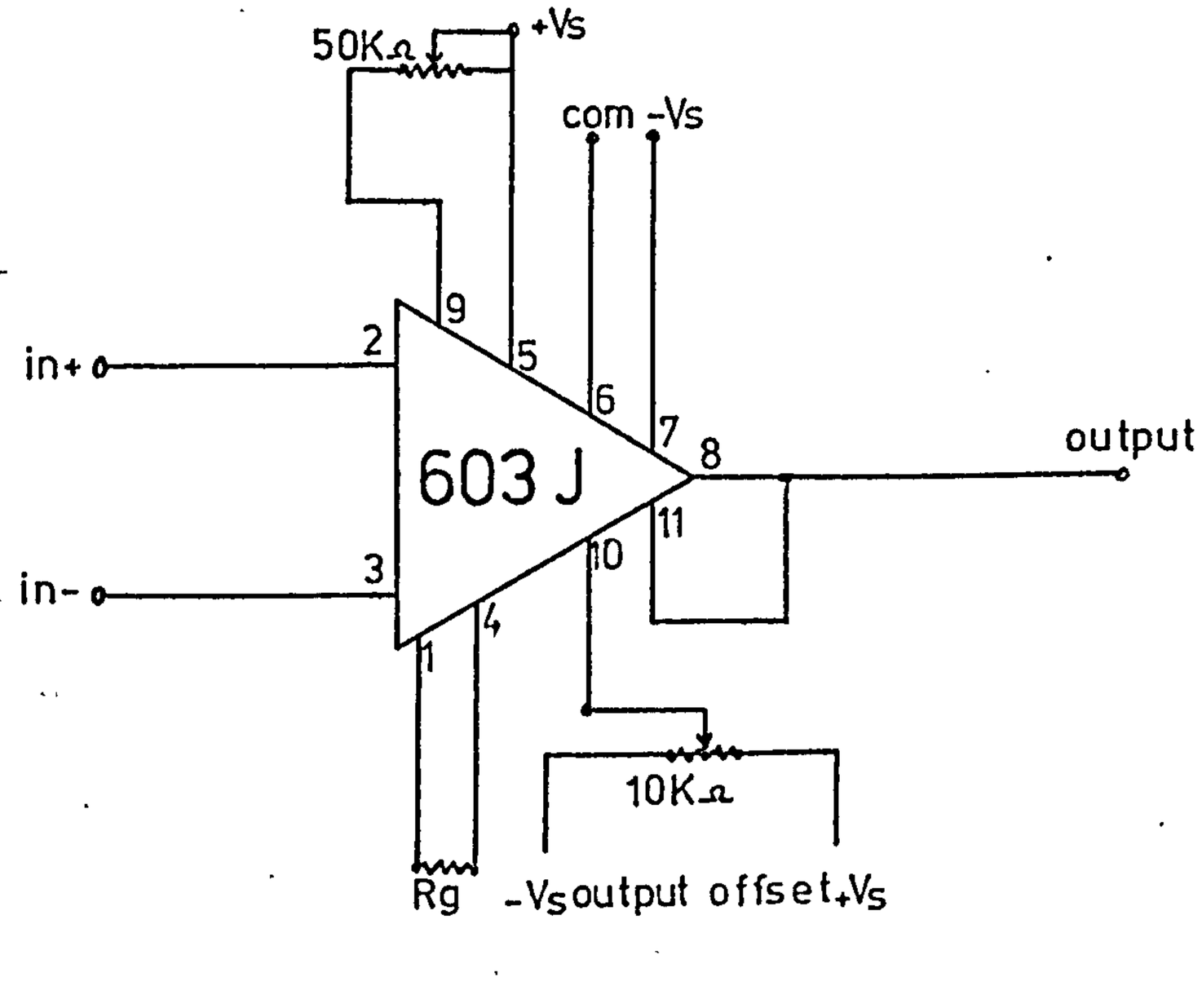
For the precise measurement of a reversible cell E.M.F. an instrument is required with adequate sensitivity and accuracy and which draws insufficient current to cause polarization. As the resistance of the selective membrane is in the order of 10^8 to 10^9 ohms, the voltmeter used must have an input impedance of at least 10^{11} ohms, thus excluding simple voltmeters. The voltmeter used in this study was a Corning E.E.L. Digital 112 research pH meter with a resolution of 0.1 mV. The input impedance of the meter is rated at $>10^{13}$ ohms.

9.2. Differential Amplifier

When an ion selective electrode is used as a reference, the voltmeter must be modified to have two high impedance inputs. Failure to carry out these modifications will result in 'open circuit' conditions being indicated by the amplifier. It was found that a low cost F.E.T. differential instrumentation amplifier made by Analog Devices (603J) was equivalent to the circuit devised by Brand and Rechnitz (101) for this application. The commercial module has a variable gain (gain = $10^5/_{Rg}$), built in output offset adjustment and an

impedance to ground of 10^{12} ohms at both input terminals. It was connected as is shown in Figure 9.2.-I. Input signals were carried by screened coaxial cables to reduce noise pickup, and the output signal was taken from terminal 8 and common (6). The power for the module was supplied by an Analog Devices 902H module with an output of 100mA.

Figure 9.2.-I Module Connections for 603J Instrumentation Amplifier



603J, Analog Devices Amplifier
numbers represent amplifier terminals

9.3. Antilog Amplifier

The potential obtained from an electrode is proportional to the log of the activity of solution to which it responds, $E \propto \ln a$. This relationship produces a potential vs. concentration graph which has a much lower gradient at higher concentration levels, introducing a larger apparent error in measurement. An antilog module has been utilized to linearize the response of the selective electrode: $E \propto a$.

The electrode voltage (E_{cell}) is given by the Nernst equation,

$$E_{cell} = E_o + S \ln a.$$

and in order for the antilogged response to be linear with respect to activity, the multiplication factor of the log term must be reduced to unity. The voltage relationship of the antilogarithmic module is given by:

$$E_{out} = E_{REF} \quad 10^{(-Ein/K)} + E_{os}$$

where E_{REF} is the reference voltage of the amplifier, E_{os} is the offset voltage, E_{in} is the input voltage , E_{out} is the output voltage and K is a scale factor. When E_{REF} is made equal to $\log^{-1}(E_o/S)$ and K is made equal to S, where E_o and S are the electrode standard potential and response slope respectively then:

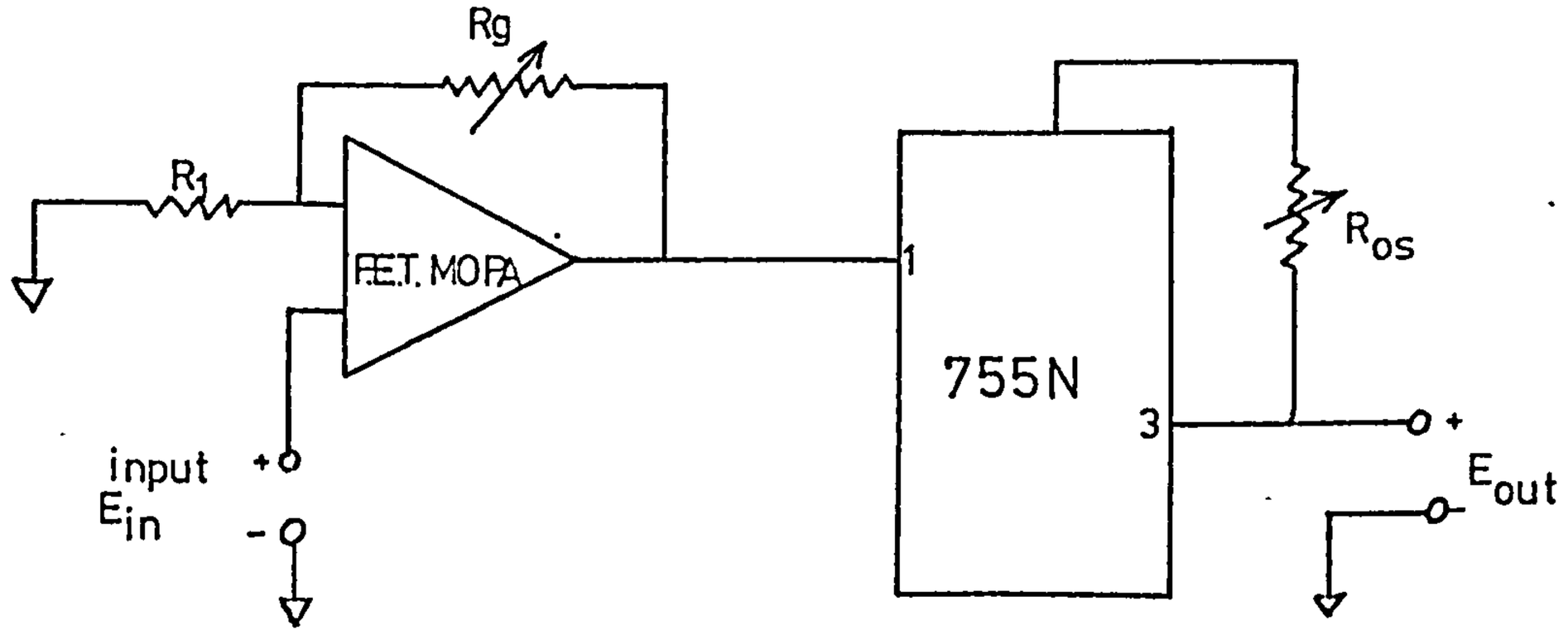
$$E_{out} = \log^{-1}(E_o/S) \times 10^{(-E_{in}/S)} + E_{os}$$

If $E_{in} = E_{cell}$, then

$$E_{out} = K'.a + E_{os}$$

The circuit diagram for this electronic arrangement is given in Figure 9.3.-I.

Figure 9.3.-I The Antilog Amplifier



$\frac{Rg}{R_1}$ = Slope Adjust ($= \frac{1}{K} - 1$). Rg = 10KΩ + 10KΩ Variable

R_1 = 2KΩ

R_{os} = 100KΩ Variable (voltage offset adjust)

755N , Analog Devices Amplifier

F.E.T. MOPA , Radio Spares Amplifier

1,3, amplifier terminals

9.4. Recorder

Signals received from all of the amplifiers were recorded directly on a "Smith Servoscribe, 115.20 Potentiometric recorder".

CHAPTER 10

P.V.C. Immobilized Electrodes, Using Commercial Liquid Ion Exchangers

10.1. The Calcium Selective Electrode

10.1.1. Preparation of the Electrode

10.1.2. Evaluation of the Membrane

10.1.3. Application of the Calcium Electrode to the Evaluation of Direct Electrode Techniques with a Non-ideal Electrode

10.2. The Nitrate Selective Electrode

10.2.1. Preparation of the Electrode

10.2.2. Evaluation of the Membrane

10.3. Conclusions

In order to evaluate electrodes made using prepared ion exchangers, it is first necessary to know the response characteristics of well tried electrode systems. In this work electrode membranes were prepared from ion exchangers that were commercially available and had been proven by continuing industrial and academic research, in order to provide a standard with which future electrodes could be compared both directly and in their application to various analytical techniques.

10.1. The Calcium Selective Electrode

10.1.1. Preparation of the Electrode

The membrane was prepared according to the optimum conditions reported by Griffiths, Moody and Thomas (82). Poly (vinyl-chloride) (0.09 gms) and 0.23 gms of Orion calcium exchanger (92-20) were mixed, dissolved in

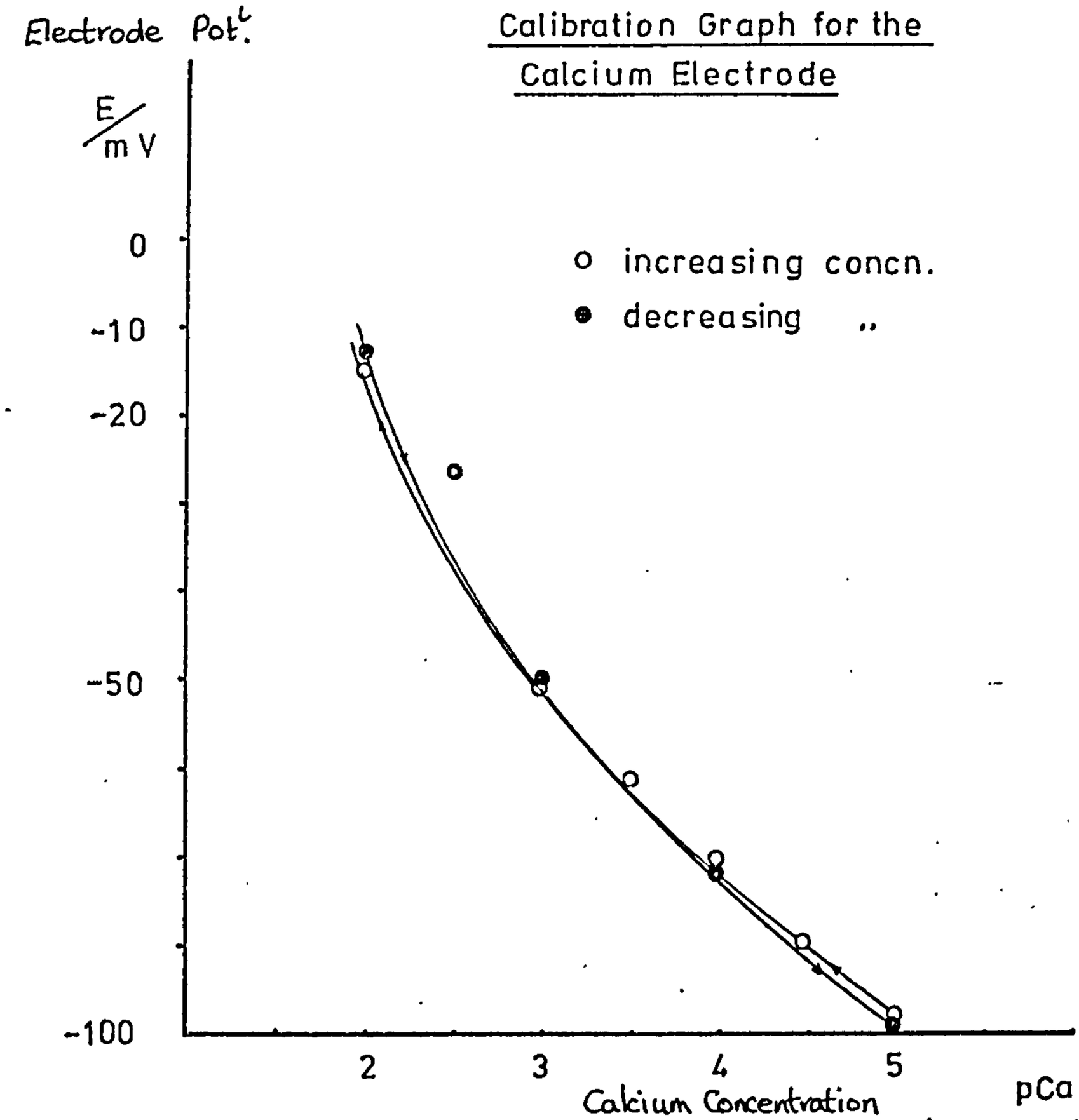
Tetrahydrofuran (T.H.F.) poured into a 1" I.D. glass ring and left covered for 48 hours for the solvent to evaporate. When the membrane was removed from the glass it was pale yellow (the colour of the exchanger) and transparent. Portions of the membrane were cut from the disc and stuck onto an electrode body with P.V.C. dissolved in T.H.F.

10.1.2. Evaluation of the Membrane

On first placing the electrode into a solution of a calcium salt the E.M.F. produced drifted rapidly and a calibration graph could not be made. After soaking in a calcium solution (10^{-3}M) for an hour the membrane became opaque, probably due to hydration of the surface, and the E.M.F. became much more stable. A test of the reproducibility of the membrane E.M.F. was made. A series of solutions were prepared and the electrode E.M.F. was measured in each while alternately increasing and decreasing the concentration of calcium. It was found that the electrode has a form of hysteresis or memory effect, the shape of the potential *vs.* concentration graph being dependent on whether it was obtained by increasing or decreasing the calcium concentrations (Figure 10.1-I). If the electrode was left for a sufficiently long period before measurement (2-3 hours) an individual potential for a specific concentration could be obtained.

The selectivity of the membrane was evaluated by method IId. The selectivity ratios found (Table 10.1-II) agree very well with those quoted for the liquid ion exchanger by Orion (94) and indicate that immobilization of the exchanger does not affect the selectivity to any great extent.

Figure 10.1.-I



Calibration Graph for the Calcium Electrode

o increasing concn.
● decreasing ,,

Table 10.1.-II Selectivity Ratios for the Calcium Electrode

K^{pot}_{ij}	j= Ni	Co	Mg	Pb	Cu	Zn
This Work 10^{-1}m	1.5×10^{-2}	3.3×10^{-3}	1.4×10^{-2}	1.1	2.3×10^{-1}	6×10^{-2}
Orion (97)			1.4×10^{-2}	0.63		3.2
Ryan (68)			4×10^{-2}	1.8	1.3×10^{-1}	3×10^{-3}

Other factors of the calcium membrane behaviour such as pH and time response have been very well characterized (67,28) and no further study of them was made.

10.1.3. Application of the Calcium Electrode to the Evaluation of Direct Electrode Techniques With a Non-ideal Electrode

In order to evaluate the common non-automated direct electrode techniques in a real analytical situation, a series of solutions was analysed for calcium content using a calcium selective electrode which had a noticeable though small potential drift and a sub-Nernstian response. All of the solutions, indicated below, were buffered to an ionic strength of I=0.06 and the potentials produced by the electrode were measured and recorded in both stirred and unstirred solutions over a period of three days. The results were then analysed by linear regression for deviation from linearity.

Solutions { Treated as standards: 2 4 6 8 10; Treated as unknowns: 4 6 } $\times 10^{-3}$m Ca

The two 'unknown' solutions were also analysed by a standard additions procedure in the presence of sodium, a known interferent (1.0M; $K^{pot}_{CaNa} = 3 \times 10^{-3}$). The results from these analyses are shown in Table 10.1.-III and indicate that with a non-ideal electrode, the most satisfactory method of analysis is that of bracketing standards. The error is always within acceptable limits and the unknown can be related to at least two known concentrations, provided that the ionic strength of all solutions remains constant.

Table 10.1-III

Solution ×10³M		Potential (-mV) unstirred					stirred					mean	mean error
"standards"	2	31·3	24·3	24·3	22·7	22·7	28·9	25·6	25·6	24·1	24·1		
"unknowns"	4	23·3	17·5	17·8	16·0	14·4	20·1	17·9	17·7	15·9	16·2	17·7	1·68
	6	17·8	13·5	12·5	12·4	9·9	15·2	13·4	12·7	11·6	11·8		
	8	13·7	12·6	9·4	8·8	6·7	12·0	10·7	8·7	8·5	8·5	9·91	1·77
	10	10·7	10·7	7·2	7·2	3·6	10·0	9·7	6·6	6·6	9·7		

Computer Analysis

"S"		29·6	19·3	24·9	22·5	26·9	27·3	23·7	27·6	25·2	22·4		
Deviation of "unknowns" from "bracket"	4	0·29	0·20	0·49	0·00	0·02	0·08	0·03	0·07	0·11	0·19	0·148	
	8	0·02	0·24	0·04	0·19	0·07	0·02	0·54	0·17	0·02	1·92	0·118	

Errors

Bracketing Standards

soln. 4 0·148mV = 1·18 %
soln. 8 0·118mV = 0·94 %

Calibration

soln. 4 1·68mV = 13·4 %
soln. 8 1·77mV = 14·2%

Standard Additions

soln. 4 = 8·5 %
soln. 8 = 4·9 %

This latter condition can be realised by addition of a high ionic strength buffer, such as T.I.S.A.B., to all solutions. The large error associated with the standard addition technique can be attributed to the variation in the slope factor. This variation is highly significant as it appears in exponential form in the calculation of results. Fortunately the variation in slope is not usually very marked, although it is frequently a characteristic of P.V.C. immobilized electrodes due to the leaching out of ion exchanger from the membrane surface.

10.2. The Nitrate Selective Electrode

10.2.1. Preparation of the Electrode

The membrane was prepared using a similar procedure to that for the calcium electrode, with Orion nitrate liquid ion exchanger (92-07). The membrane, which was pale brown becoming darker with increased exposure to air and light, was incorporated into the electrode end cap with an internal filling solution of 1:1 KCl, KNO_3,(10^{-1}M).

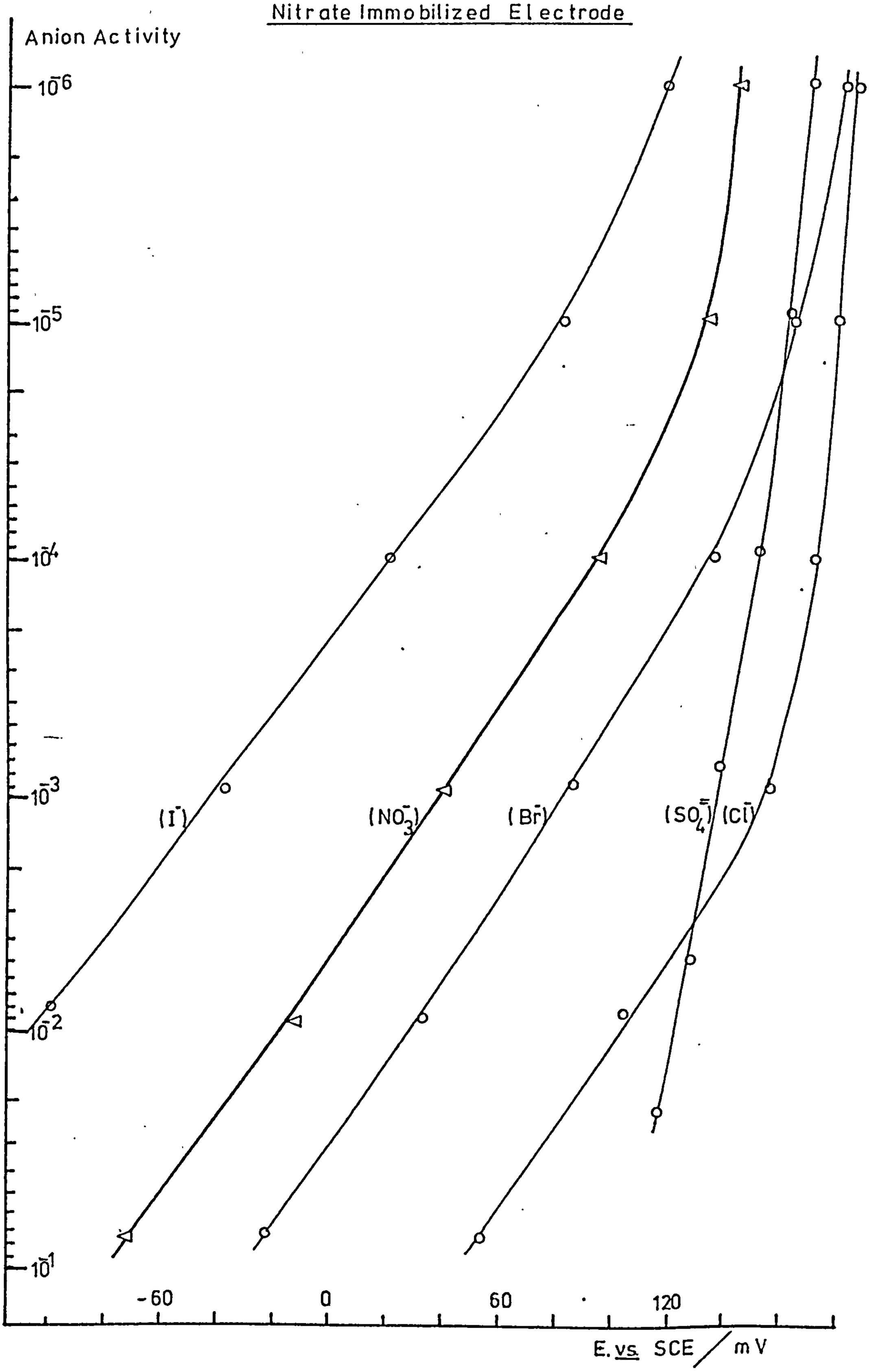
10.2.2. Evaluation of the Membrane

The membrane was soaked in 10^{-3}MKNO_3 for an hour and then calibrated in solutions of potassium nitrate. The electrode showed some hysteresis although it was less evident than for the calcium electrode. The mean values obtained for the calibration are shown in Figure 10.2.-I.

Solutions of various anions were made to test the selectivity of the electrode, using a separate solutions method, calibration graphs for which are plotted in Figure 10.2.-I. The selectivity ratios calculated by methods Ia and Ib are given in Table 10.2.-II.

Figure 10.2.-I

Calibration and Selectivity Evaluation of the Nitrate Immobilized Electrode



An evaluation of the dynamic time response of the nitrate membrane electrode was made. 1 ml of 10^{-2}M potassium nitrate was added to 10 mls of 10^{-4}M solution with a syringe. The potential of the membrane was recorded as the concentration changed from 10^{-4} to 10^{-3} molar, and a typical plot is shown in Figure 10.2.III. The value of t_{95} (time to reach 95% of the equilibrium potential) was in the order of 4 seconds, although the shape of the curve was found to be very sensitive to the way in which the concentrated solution was injected.

10.3. Conclusion

Ion selective electrodes made by P.V.C. immobilization of commercial liquid ion exchangers are easy and quick to prepare as well as being cheap and effective analytical tools. The time response of the membranes, although two orders of magnitude slower than their solid state counterparts, is still adequate in most cases for their application to continuous methods of analysis and as endpoint sensors in automated titrations.

Table 10.2.–II

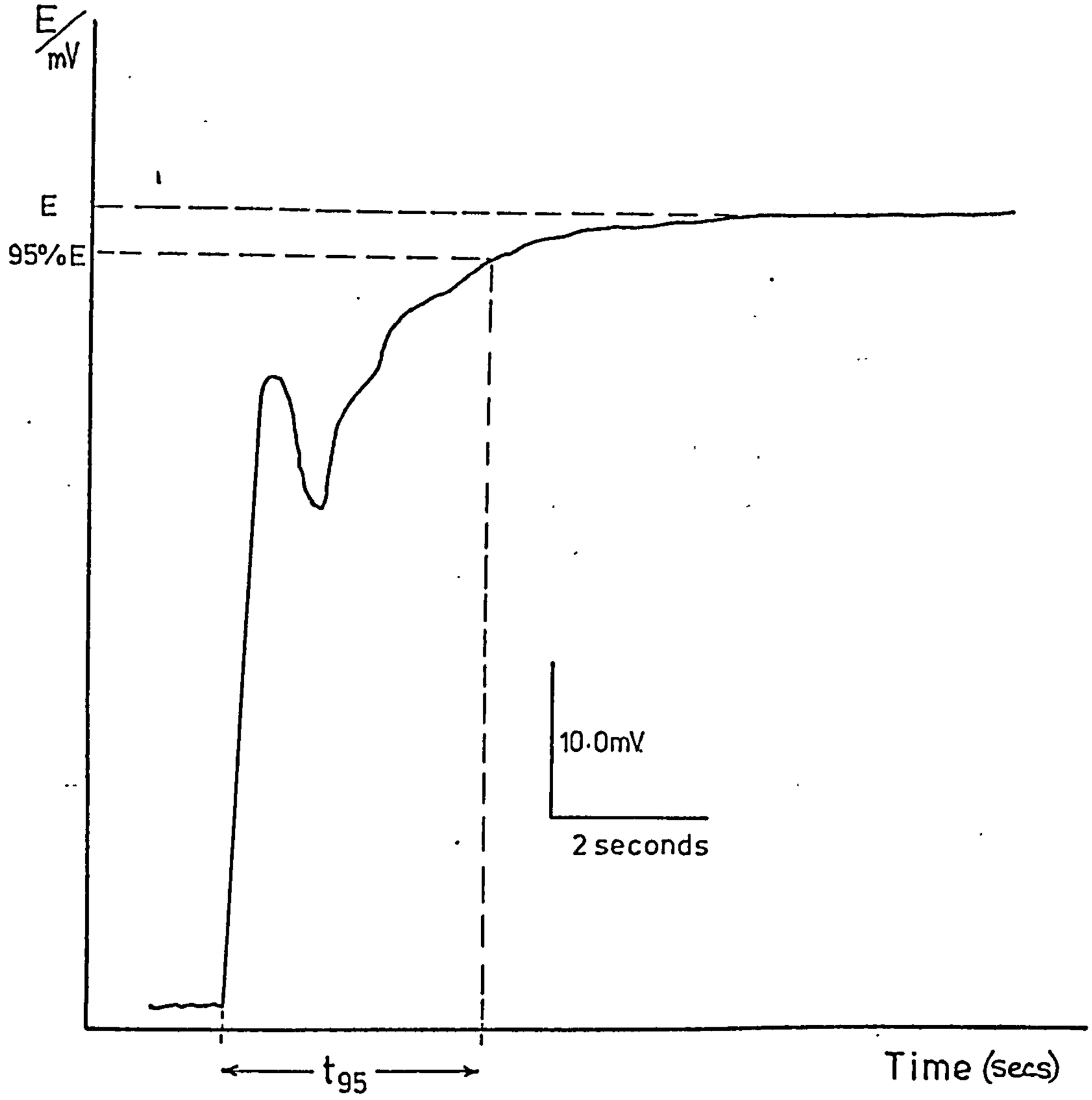
Selectivity Ratios of the Nitrate Electrode

NO_3^- Concn.	10^{-2}		10^{-3}		10^{-4}		Orion
Method I	a	b	a	b	a	b	
ION							
I^-	21.3	34.0	18.2	25.9	20.6	16.1	20.0
Br^-	0.16	0.15	0.15	0.16	0.16	0.19	0.1
Cl^-		8.2×10^{-3}		1.2×10^{-2}	6.9×10^{-3}	4.7×10^{-2}	4×10^{-3}
SO_4^{2-}				5.7×10^{3}			3×10^{5}

Figure 10.2-III

Time Response of the Immobilized Nitrate Electrode

($\bar{1}0^4 \longrightarrow \bar{1}0^3$ M)



t_{95} = 3.4 seconds

CHAPTER 11

The P.V.C. Based Water Hardness Electrode

11.1. Preparation of the Membrane

11.2. Evaluation of the Membrane

11.3. Application of the Water Hardness Electrode As An Endpoint Sensor for Titrations

11.4. Conclusions

Water hardness is caused by the dissolved salts of calcium, magnesium and iron. In industry the hardness of the water can be very important, for example when water is used for cooling and steam generation any 'temporary' hardness will cause a carbonate deposit in the pipelines. Both temporary and permanent hardness, the latter caused by the sulphates of the metals, will give insoluble scum (salts of the fatty acids) when soap is used to wash materials such as wool in processing plants. In order to evaluate the effectiveness of any method of removal, an efficient means of measuring the quantity of metal ion in solution must be available. Titrations of water hardness with soap solutions to an audible endpoint have been used, but they require considerable expertise to ensure accuracy. E.D.T.A. is also used as a titrant, with Solochrome Black as the indicator, but in this case magnesium must be present to ensure a sharp endpoint and the cations of copper, cobalt or nickel will cause inhibition of the indicator. The calcium ion selective electrode can be used to measure the amount of calcium in solution by both direct potentiometric and titration techniques but since it was developed to be

selective over magnesium, it is of little use in measuring this ion. The calcium electrode however has been used as an endpoint sensor in the titration of calcium and magnesium together with an E.D.T.A. titrant (163).

In order to measure the hardness of water by a direct potentiometric method, utilizing the speed of selective electrodes, a membrane responsive to all divalent metals must be developed. This membrane must be not only responsive to all of these ions but also be equally selective to them. The water hardness electrode developed by Orion is based on calcium didecyl phosphate dissolved in decanol (94) which is quoted to be equally selective to both calcium and magnesium. As the electrode is responsive to most divalent ions it can also be used in such applications as the titration of many ions with precipitants,for example barium with sulphate. The major disadvantages of the electrode are that in the liquid form it is difficult to prepare, uses a large amount of ion exchanger, and has a fragile membrane. Immobilization of the ion exchanger in a P.V.C. matrix should alleviate most of these problems and provide a convenient means of measuring water hardness by both direct potentiometric and titration analysis, with the added advantage of its possible incorporation into automated methods.

11.1. Preparation of the Membrane

The ion exchanger was prepared by dissolving calcium didecylphosphate ($Ca(C_{10}H_{21})_2PO_2$) in decanol. 0.154g of the calcium salt were added to 6 ml of decanol.

The mixture was heated to 100°C in a water bath to dissolve the salt and allowed to cool to form a viscous translucent liquid. A more dilute exchanger solution was also prepared (10^{-2}Molar).

The membrane was cast by immobilizing the exchanger in P.V.C. using four separate mixtures of P.V.C./exchanger.

Membrane	A	B	C	D
P.V.C.	0.1g	0.1g	0.1g	0.1g
Exchanger	0.1	0.2	0.4	0.4 (10^{-2}M)

The decanol solvent is not a plasticiser for P.V.C. and the membrane produced is brittle and wax-like. Addition of di-n-octyl sebacate (0.1%) plasticises the P.V.C. and the membrane becomes flexible and rugged.

In all cases, after leaving the membranes for 48 hours to dry the solvent (decanol) was excluded from the surface and formed a layer over the membrane, which was removed with tissue paper before it was stuck to an electrode end cap.

11.2 Evaluation of the Membranes.

Membranes A and B rapidly became white and opaque in aqueous solutions with loss of the decanol. They initially showed a cationic response but the potential drifted rapidly and the membranes soon became non selective. The potential produced by membrane D drifted excessively and quickly became permselective.

Membrane C was prepared and calibrated in solutions of calcium nitrate. The calibration graph shown in Figure 11.2.-I is typical for divalent cations, with a

Calibration Graph of the Water Hardness Electrode

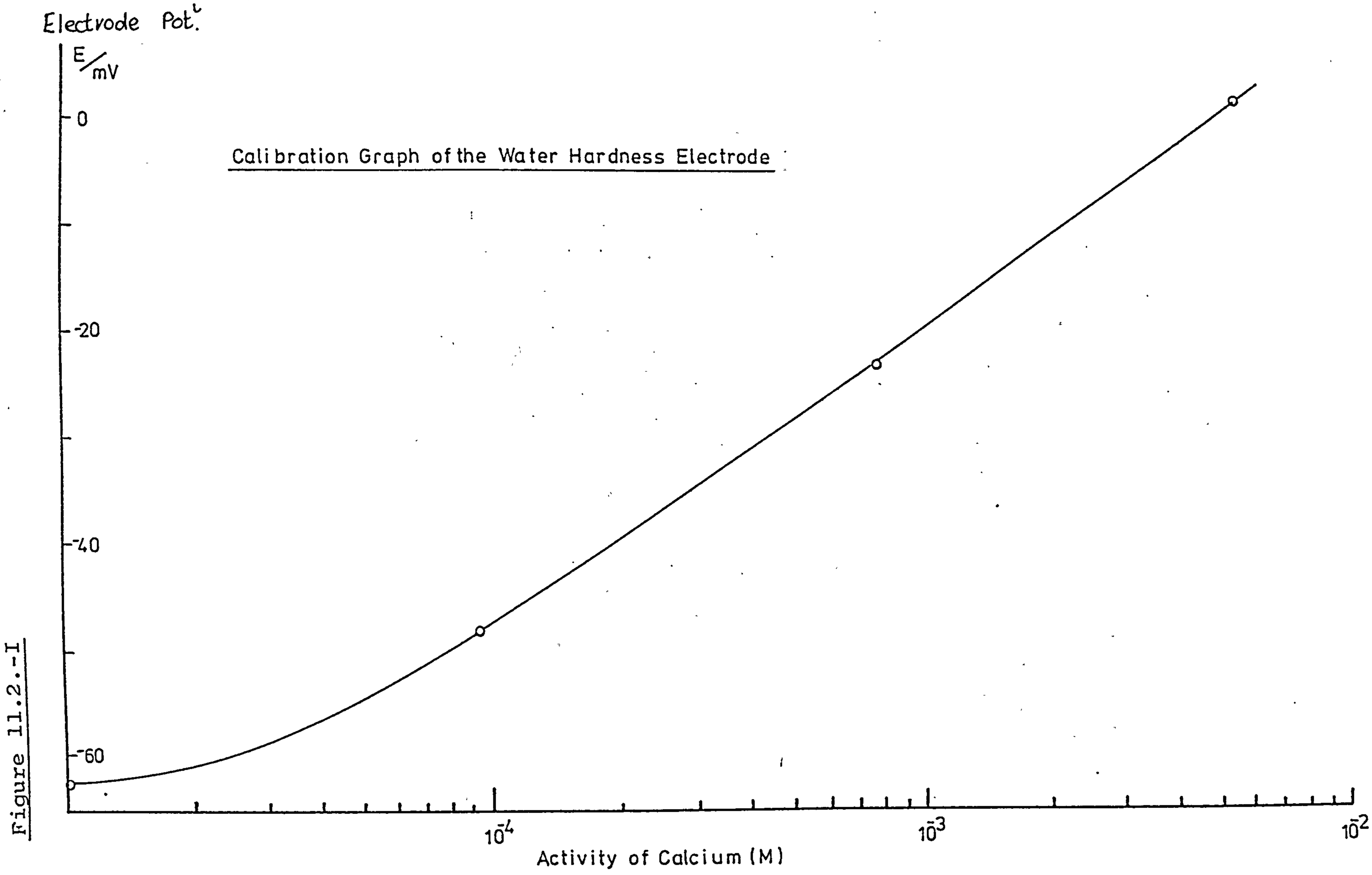


Figure 11.2.-I

slope of 28mV per decade activity at 25°C. After continuous use over a period of 48 hours the slope had decreased to 25mV and remained at this value for a further 10 days. Calibration of the electrode in solutions of constant ionic strength showed a response of 25 mV a decade which was linear from 10^{-2} to 10^{-4} molar.

The dynamic time response of the membrane was studied for a concentration change from 10^{-3} to 10^{-2} molar (Figure 11.2.II). The mean value of t_{95} was 8±1 seconds, showing that the membrane tended to respond more slowly than the immobilized calcium exchanger.

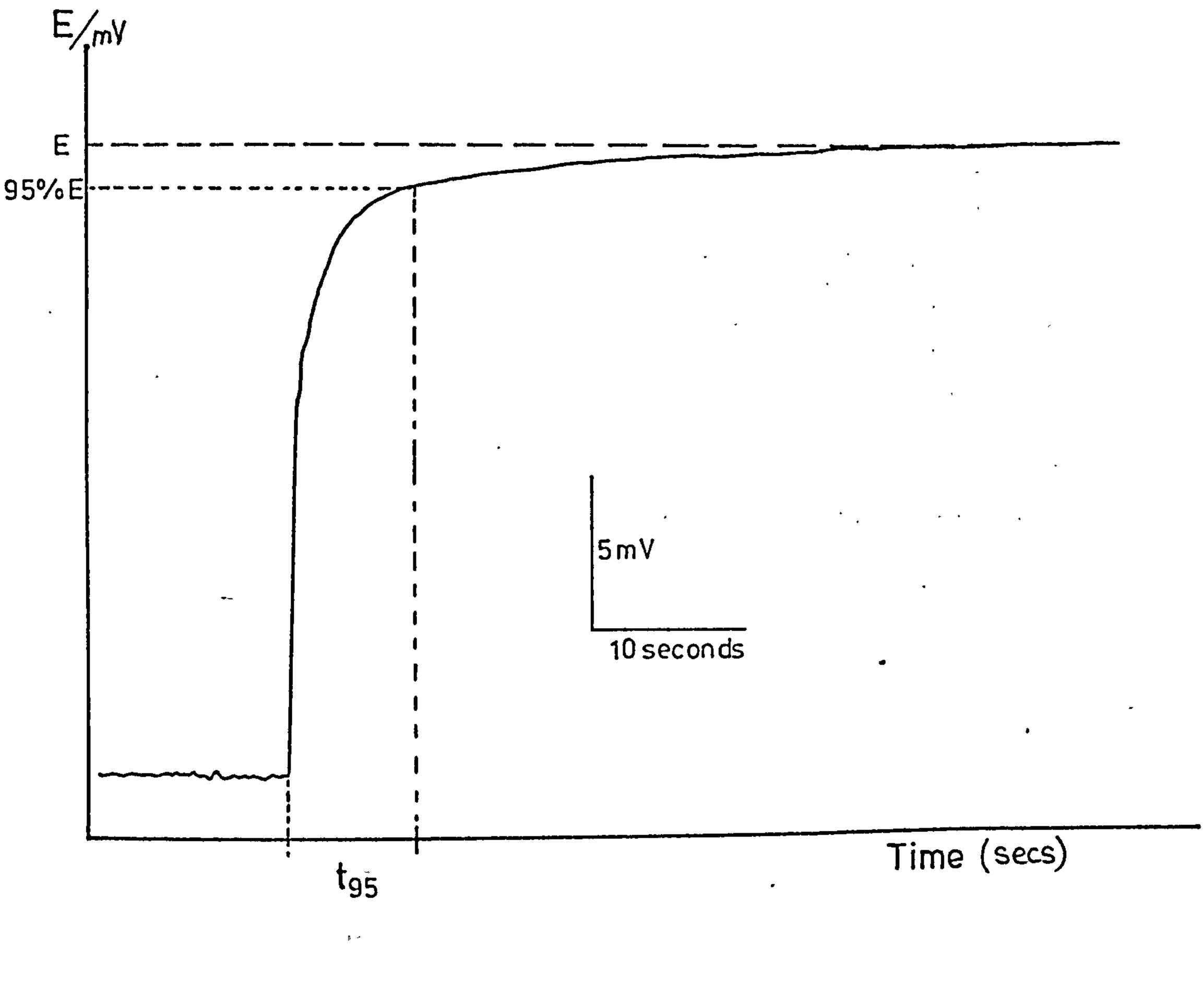
In order to respond effectively as a water hardness electrode the membrane should respond equivalently to both calcium and magnesium and preferably to other cations also. In order to test this a series of solutions were made up with varying concentrations of calcium and other ions while maintaining a constant cation activity. The potentials provided by the water hardness membrane were measured and plotted, as can be seen in Figure 11.2.-III. From these results it can be seen that although the membrane does not significantly differentiate between calcium and magnesium, both lead and zinc have a selectivity coefficient greater than unity.

The selectivity coefficients for these ions were evaluated in mixed solutions by method IIb, and the results are shown in Table 11.2.IV with the values published by Orion for their liquid exchanger water hardness electrode.

Figure 11.2.-II

Time Response of the Water Hardness Electrode

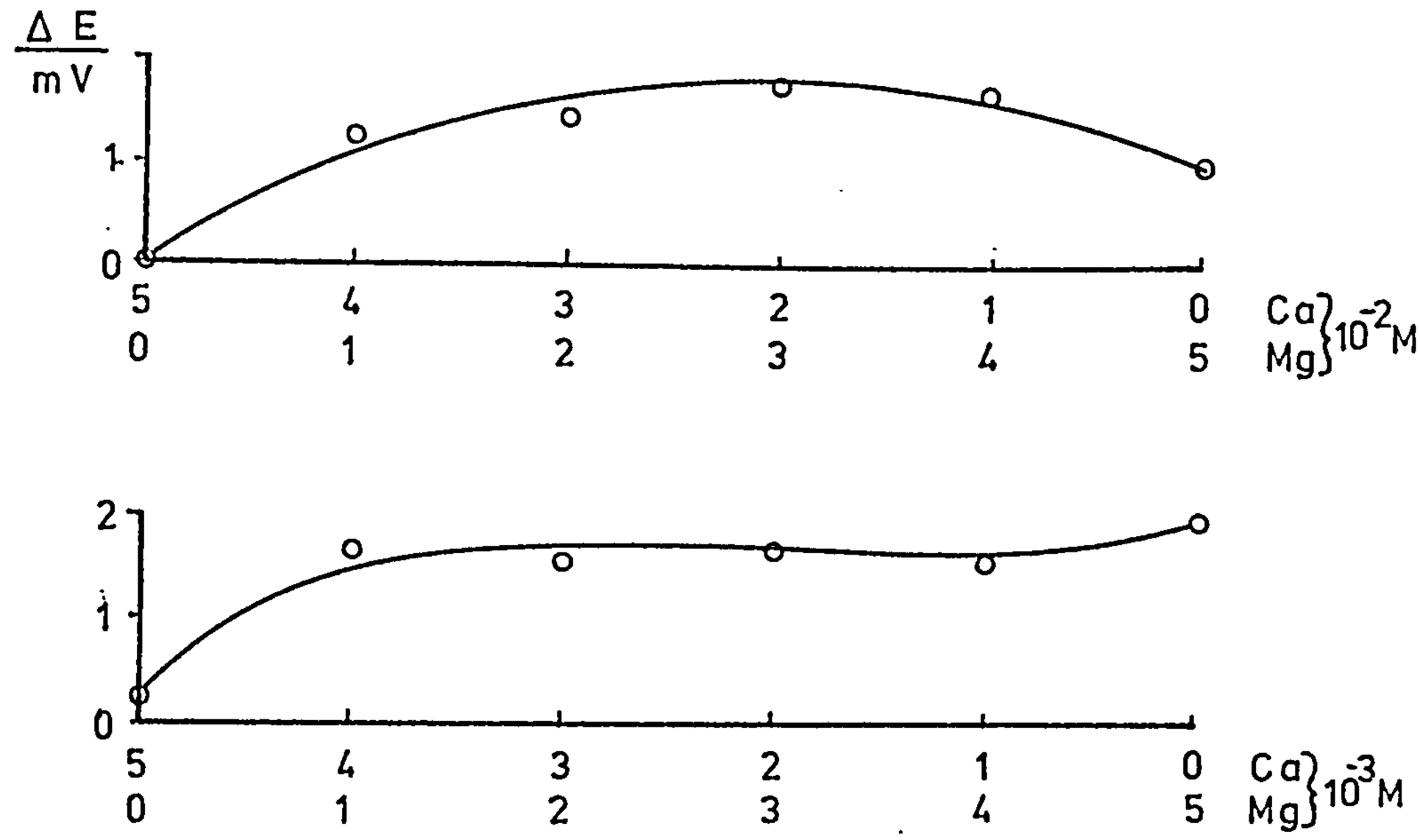
($10^{-3} \longrightarrow 10^{-2}$ M)



t_{95} = 8±1 seconds

Figure 11.2.-III

Performance of the Water Hardness Electrode in Mixed Solutions



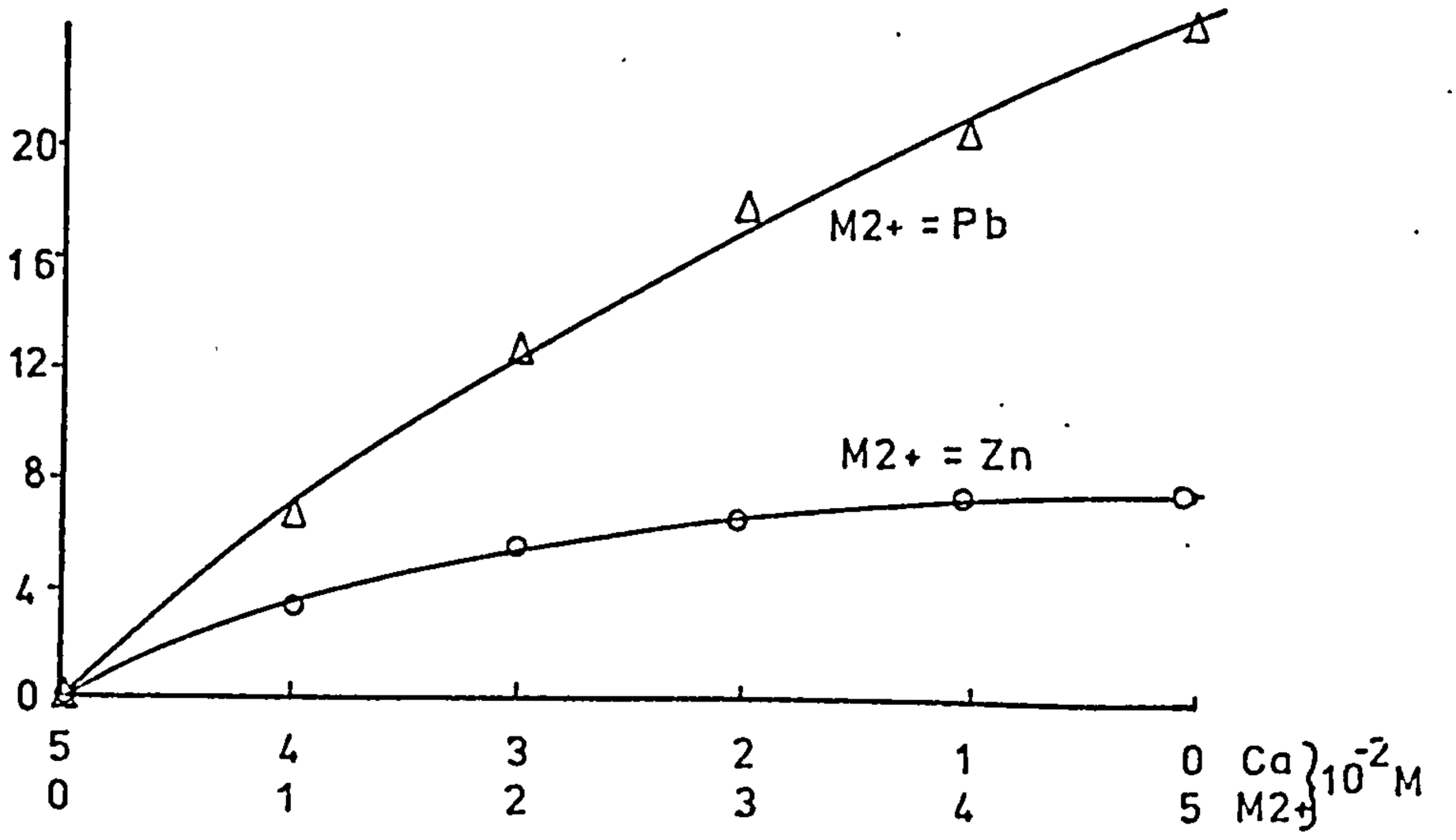


Table 11.2-IV K^{pot}_{CaM}

Ion (10^{-1}M)	Zn	Pb	Mg	Ba
Orion	3.5	—	—	0.94
This Work (IIb)	3.3	20	91	1.3

11.3. Application of the Water Hardness Electrode As An Endpoint Sensor For Titrations

The water hardness electrode was used as an end-point monitor in a calcium/E.D.T.A. titration. The solutions were kept at pH 9.7 with a glycine buffer and both E.D.T.A. and calcium were used as the titrant. The potential plots for these titrations are given in Figure 11.3.-I, showing clearly defined endpoints.

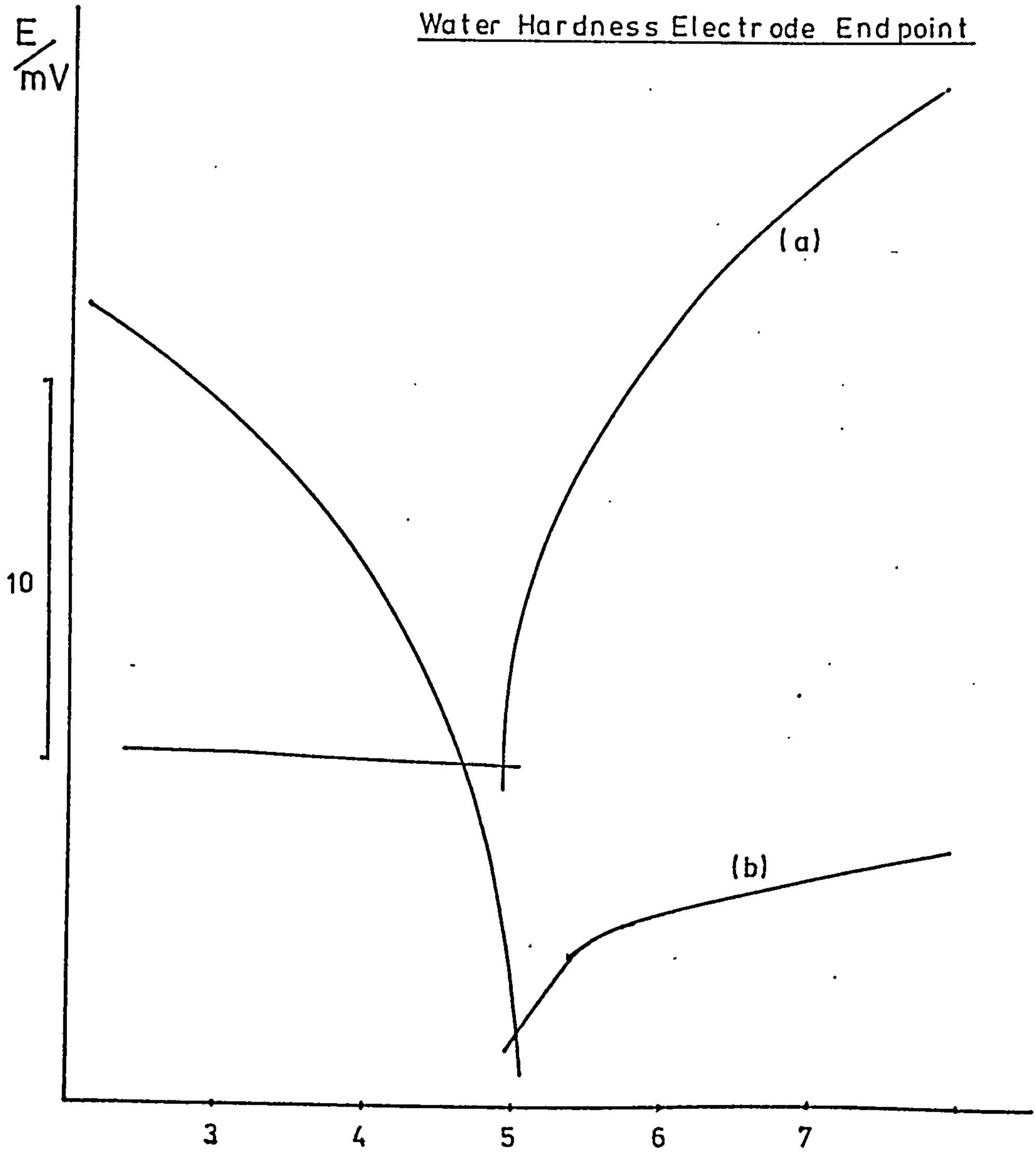
Mixed solutions of calcium and magnesium salts were prepared and titrated with E.D.T.A. using a pH 10 NH_3/NH_4Cl buffer. The titration curves, examples of which are shown in Figure 11.3.-II, were identical indicating that the electrode will give a sharp response at the end-point regardless of whether there is magnesium present or not in the sample (in contrast with the colourimetric methods of analysis).

11.4. Conclusions

Immobilization of the liquid exchanger for the water hardness electrode does not interfere with the selectivity of the membrane, and the P.V.C. matrix provides a rugged electrode with a life time of several weeks in normal use. The electrode does not significantly differentiate between divalent metals, and because of this

can successfully be used to measure total calcium and magnesium hardness in water. The effectiveness of the membrane as a sensor in potentiometric titrations will enable its use in automated titration applications as well as in direct potentiometic systems.

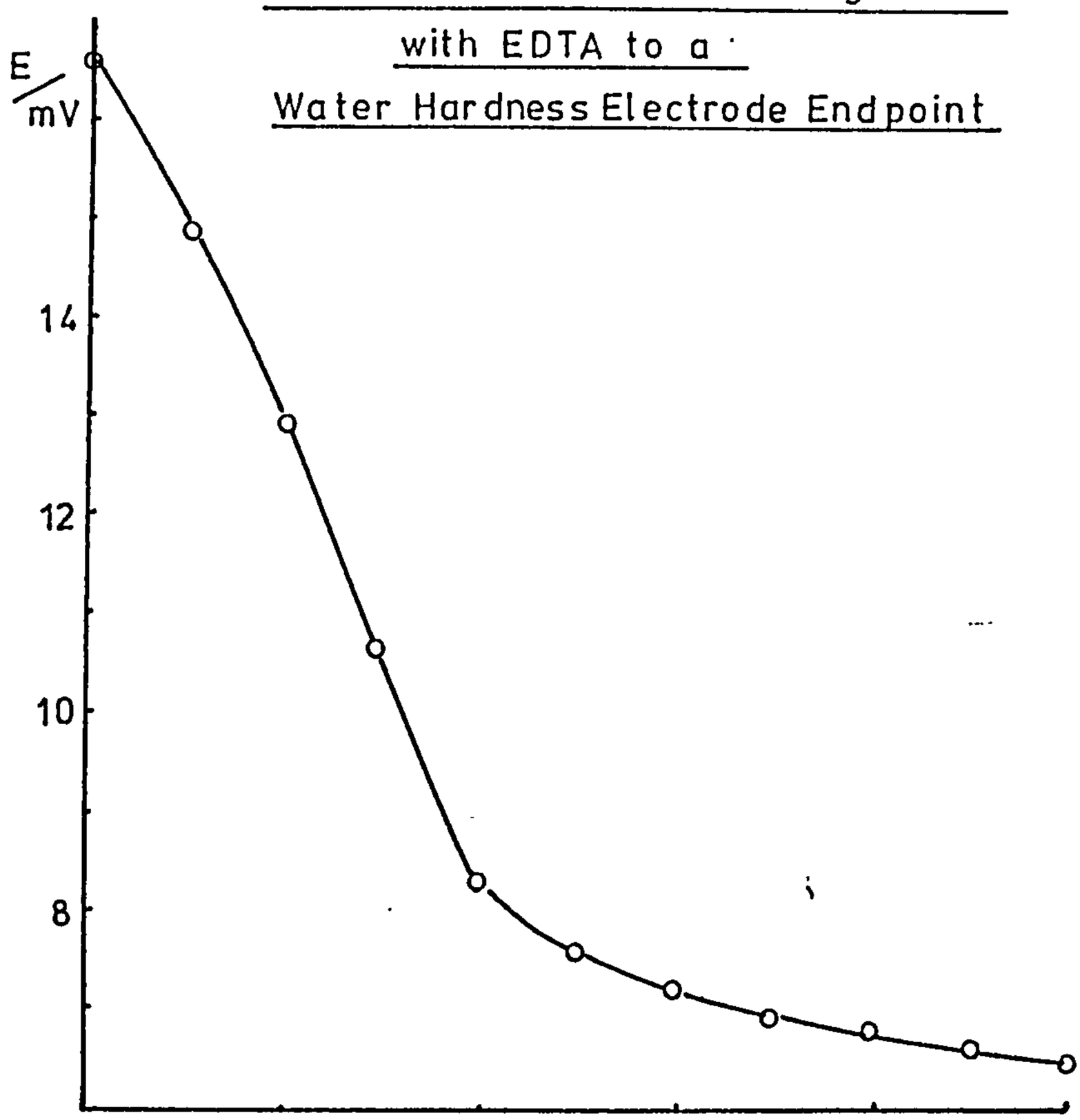
Figure 11.3.-I Titrations of Calcium and E.D.T.A. to a Water Hardness Electrode Endpoint



Glycine buffer (pH 9.7)
(a) Ca titrant
(b) E.D.T.A. titrant

Figure 11.3-II

Titrations of Calcium and Magnesium with EDTA to a Water Hardness Electrode Endpoint



Titrant 0·4M E.D.T.A.

All Points ±0·1mV for Solutions a,b,c are Coincident

(a) 1.6×10^{-2}M. Ca	(b) 8×10^{-3} M. Ca + 8×10^{-3} M. Mg	(c) 1.6×10^{-2} M. Mg

CHAPTER 12

The Development And Application Of An Immobilized Antibiotic Lead Sensitive Ion Selective Electrode

12.1. Currently Available Lead Electrodes

12.2. The Antibiotic (Virginiamycin)

12.3. Preparation Of The Membrane

12.4. Initial Study Of Electrode Response

12.4.1. Permselectivity

12.4.2. Specificity

12.4.3. Sensitivity

12.4.4. Selectivity

12.4.5. pH Response

12.4.6. Time Response

12.5. The Effect Of Variations In The Antibiotic Membrane

12.5.1. Solvent Changes

12.5.2. Antibiotic Change

12.6. Application Of The Virginiamycin (M) Electrode

12.6.1. Lead/Sulphate Titration

12.6.2. Lead/Dichromate Titration

12.6.3. Lead/E.D.T.A. Titration

12.7. Conclusion

12.1. Currently Available Lead Electrodes

The majority of lead sensitive electrodes available are based on a mixture of silver and lead sulphides (38,23), in which the electrode responds to lead through interaction with both sulphide and silver. The sulphide mixture is difficult to press into a pellet however and the necessary conditions are given by Czaban and Rechnitz (38).

This difficulty has been overcome by incorporating the ion exchanger into a siliconerubber (34) or P.V.C. matrix (102) and by coating the end of a hydrophobitised graphite bar as in the Selectrode (43,44). The membrane may also be made in ceramic form by heating the mixture at a high temperature for a prolonged time (103). All of these electrodes suffer the same interferences (notably Cu^{2+}, Ag^{+} and Hg^{2+}) and have a linear range varying from 3 to 5 decades of activity. The sensitivity of the sulphide based membranes decreases in acidic media due to the formation of hydrogen sulphide. An improvement in this effect has been reported with the electrodes based on lead selenide and lead telluride (104). These electrode responses are linear over 6 decades of activity but the same ions are noted as interferents.

A lead electrode marketed by Orion based on thioacids (105) has also been described and evaluated (106), but only had a linear range of two decades and the range of interferences was more serious than those for the solid state electrodes. In addition a patent has been granted to Miles Laboratories (107) for a lead electrode based on P.V.C. immobilized solutions of chelating agents, such as thenoyltrifluoroacetone and diphenylthiocarbazone. The sensitivity and selectivity are not quoted but on theoretical grounds this membrane configuration would not be expected to be efficient.

It was hoped that the replacement of chelating agents and precipitate based ion-exchangers by antibiotics with a favourable selectivity towards lead would provide the means to make an electrode with the properties necessary for an efficient analytical tool.

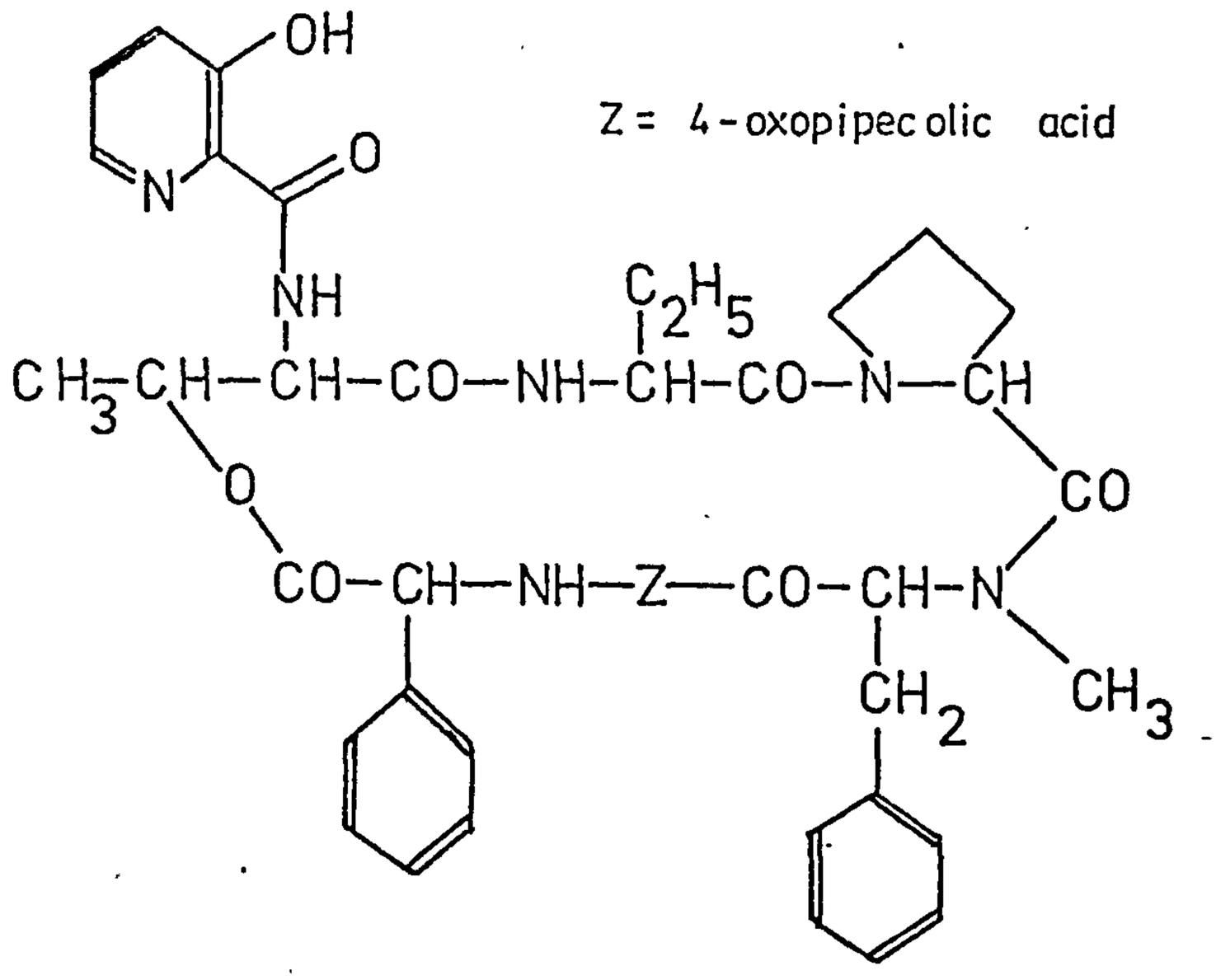
12.2. The Antibiotic (Virginiamycin)

The antibiotic used in this study is a readily available compound. Formerly known by its trade name Staphylomycin, 'Virginiamycin' is a common animal feedstuffs additive. It is a mixture of two antibiotic components, known as factors M and S (108). Factor 'S' is a cyclic depsipeptide, (C_{43} H_{49} N_7 O_{10}), a similar compound to the well known antibiotic valinomycin, and is active mainly against Bacillus subtilis. The structure of Factor 'S' is given in Figure 12.2.-I. Factor 'M' is a macrocylic lactone (C_{28} H_{35} N_3 O_7) with the structure shown in Figure 12.2.-II. It is active mainly against Micrococcus aureus. The analysis of a sample of virginiamycin has shown that the percentages of other minor virginiamycin factors are very low.

12.3. Preparation Of The Membrane

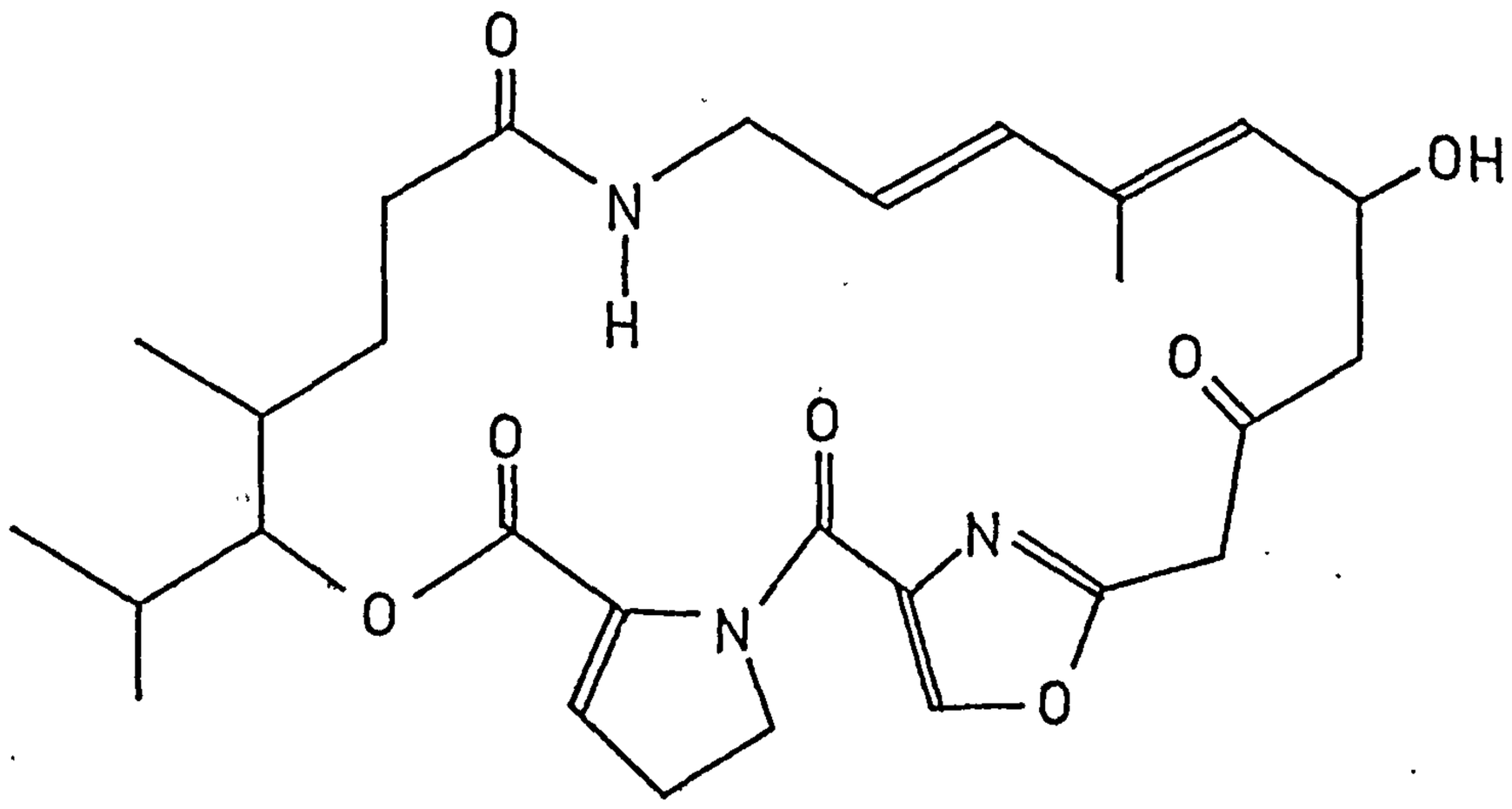
The ion exchanger was prepared by dissolving 2mg of virginiamycin (M) in 1 ml. of solvent. The solvent is a multicomponent mixture containing a plasticiser for the P.V.C. matrix. The exchanger (0.3g) was mixed with low molecular weight P.V.C. (B.D.H., 0.1g), and sufficient tetrahydrofuran (T.H.F.) was added to make a viscous solution. The mixture was poured into a 1" I.D. glass ring on a glass plate and left covered for 48 hours. The membrane formed was ~0.3 mm thick, rubbery, transparent and colourless. Small discs, cut with a cork borer, were stuck onto a P.V.C. tube with a solution of P.V.C. in T.H.F. Internal contact with the membrane was accomplished by means of a silver, silver chloride

Figure 12.2-I Virginiamycin Factor S



Z = 4-oxopipecolic acid

Figure 12.2-II Virginiamycin Factor M



wire and a filling solution. As an alternative design a solid contact may be made by pressing a carbon rod (spectrographic grade) against the internal surface of the membrane, thus dispensing with the internal solution.

12.4. Initial Study Of Electrode Response

12.4.1. Permselectivity

To find out if the membrane responded to anions or cations, the electrode potentials in several different unbuffered solutions of various salts were measured. An increase in potential with increasing concentration indicates that the membrane responds to cations, according to the Nernst equation:

$$E = E^{o} + \frac{RT}{nF} \ln(a)$$ n(positive) for cations, (negative) for anions.

Table 12.4.-I

Salt Solution	$\partial E / \partial \ln(a)$
NaCl	+ve
K Cl	+ve
NH_4Cl	+ve
$Ca(NO_3)_2$	+ve
Ni SO_4	+ve

The results shown in Table 12.4.-I indicate that the membrane responds to cations in solution.

12.4.2. Specificity

The next stage in the membrane evaluation was to investigate the membrane specificity. If the membrane selectively responds to any cation its potential will change with an increase in the cation concentration, even in the

presence of a base electrolyte. To study this effect, several additions of various salt solutions (1ml, 10^{-1}M) were made to an ionic buffer and the potential was monitored. The results, shown in Figure 12.4.-II indicate that the electrode has a very poor response to most cations, the negative slope indicating a dilution of the ionic buffer; whereas the response towards lead ions is far greater than the response to any other cation, indicating that the membrane might be used as a lead selective ion electrode.

12.4.3. Sensitivity

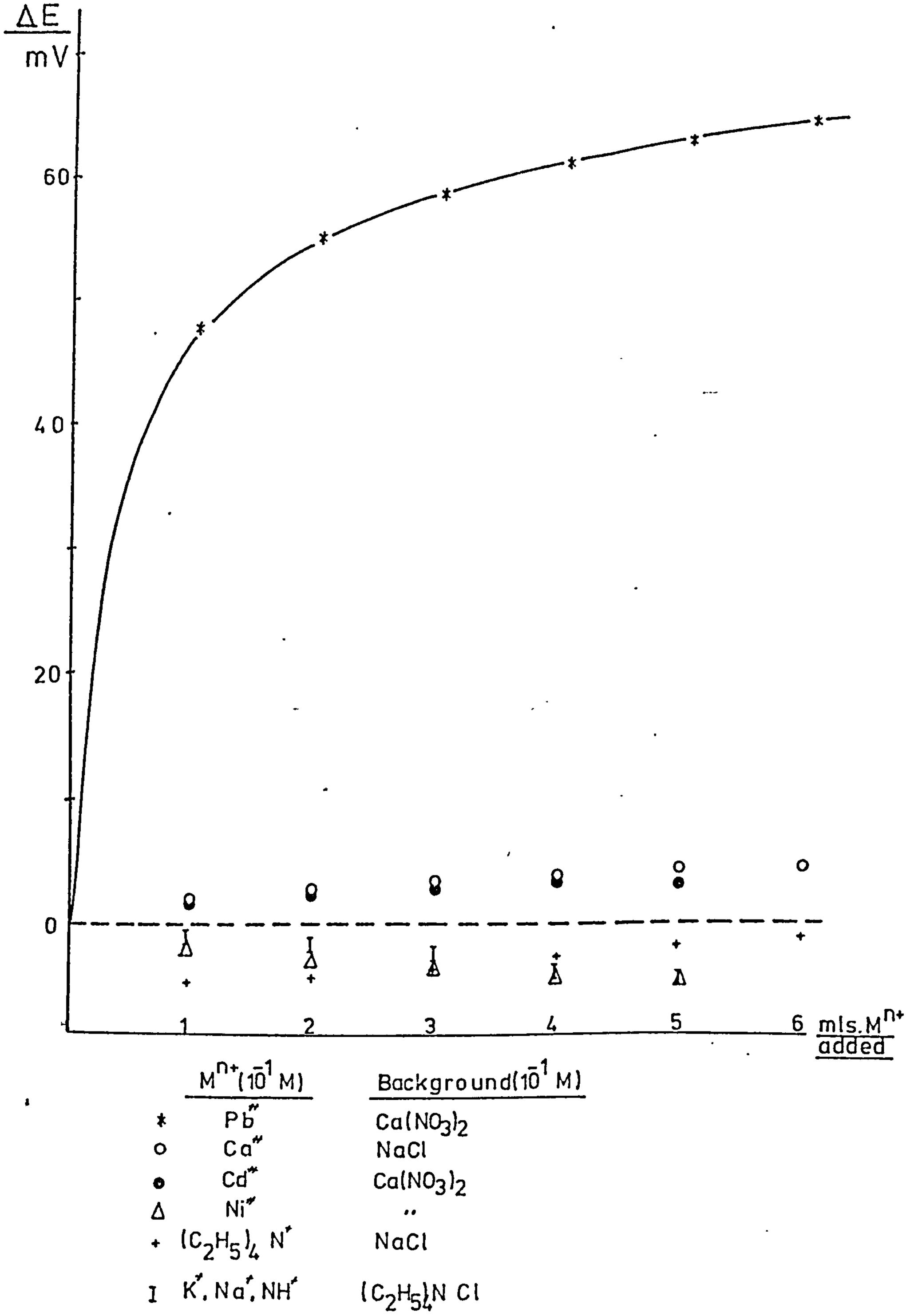
To investigate the response of the membrane as a lead selective electrode the sensor was calibrated in a series of lead nitrate solutions, buffered ionically with potassium nitrate and adjusted to pH4 with nitric acid. The internal filling solution used during the calibration was KCl (10^{-1}M) saturated with $PbCl_2$. The graph obtained (Figure 12.4.-III) shows that the membrane has an exceptionally good response towards lead for an immobilized liquid membrane electrode, being almost Nernstian over the concentration range 10^{-1} M to 4 x 10^{-6}M. Repeated calibrations in both increasing and decreasing concentrations showed that the electrode suffered very little from 'memory' of the previous solution.

12.4.4. Selectivity

The methods available for the determination of membrane selectivity have been described in Section II,4.2. Using the separate solution methods (Ia,b) the calculated activities of several ions at various concentrations were

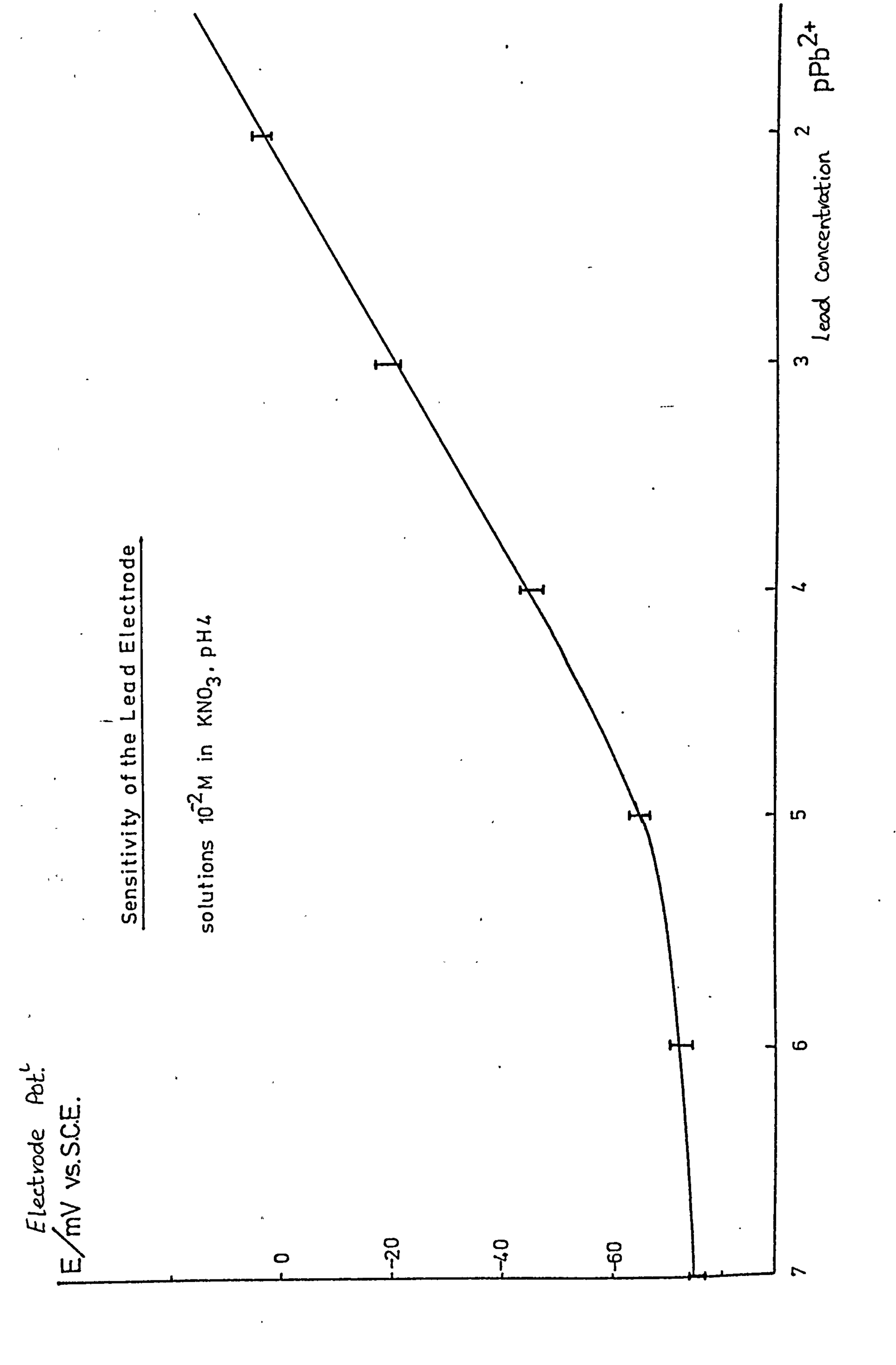
Figure 12.4.-II

Lead Electrode Specificity



	M^{n+} (10^{-1} M)	Background (10^{-1} M)
*	Pb^{++}	$Ca(NO_3)_2$
o	Ca^{++}	NaCl
⊙	Cd^{++}	$Ca(NO_3)_2$
Δ	Ni^{++}	"
+	$(C_2H_5)_4 N^+$	NaCl
I	K^+, Na^+, NH^+	$(C_2H_5)_4N$ Cl

Figure 12.4.-III



plotted vs. electrode potential. The resulting curve (Figure 12.4.-IV) shows a good separation between lead and other cations. It is also interesting to note that the gradients of the interferent curves are smaller than that for the lead ion, introducing an error into the results for the separate solution methods of selectivity analysis which are given in Table 12.4.-V.

The value of a selectivity ratio in pure solutions can have no real value as the membranes will be needed for analysis in real samples of mixed ions. Method IIa was used for the zinc and copper interferents to provide a comparison of the various techniques and the results for this method, with the results for method IId are given in Table 12.4.-V. In method IId, six, 1.0 ml additions of 10^{-1}M lead nitrate were added to 25 ml samples of interferent solution ionically buffered (to 10^{-1}M) with potassium nitrate. The resulting electrode potentials, measured after each increment, were processed by the computer programme ADDFIT (88) to give an apparent lead concentration C_A. When this value was divided by the actual interferent concentration, C_J, the selectivity coefficient was obtained, $K^{pot}_{PbJ} = C_A/C_J$. The selectivity coefficients for a commercial Orion lead electrode were computed by the same method for comparison.

12.4.5. pH Response

The virginiamycin membrane's response to pH in the presence of lead was investigated. The lead electrode, a glass pH combination electrode and a calomel reference

Figure 12.4.-IV

Selectivity Evaluation of the Lead Electrode

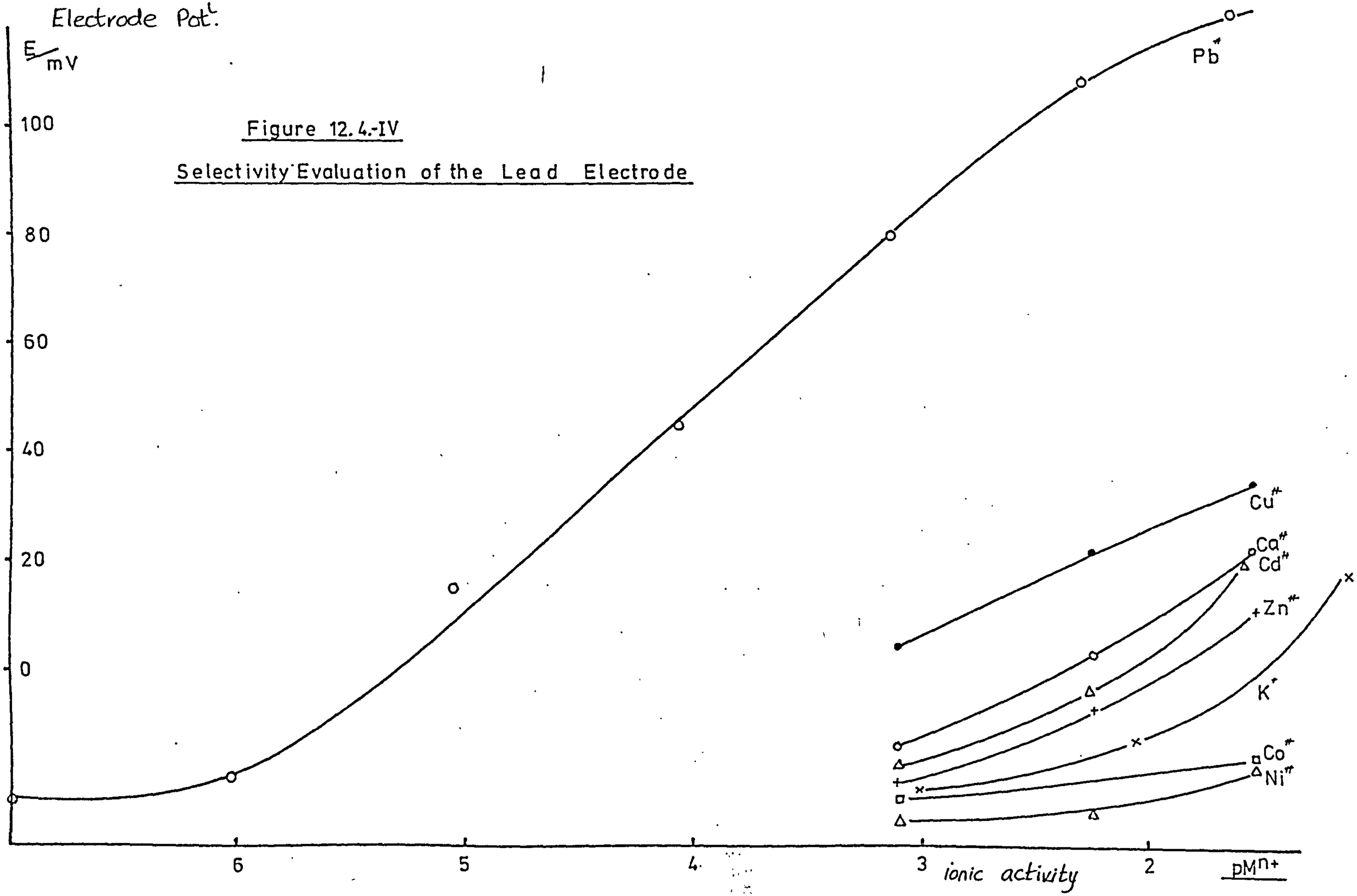


Table 12.4.-V

Results of Selectivity Ratio (K^{pot}_{PbM}) Analysis

Method of Analysis		Ia		Ib		IIa	IId			
Ionic Concentration (M)		10^{-2}	10^{-3}	10^{-2}	10^{-3}	10^{-2}	10^{-1}	10^{-2}	10^{-3}	10^{-4}
Virginiamycin	Ca^{++}	2.4×10^{-4}	1.1×10^{-3}	8.5×10^{-4}	1.8×10^{-3}		3.8×10^{-4}	1.1×10^{-3}	1.8×10^{-2}	1.3×10^{-1}
	Cd^{++}	1.6×10^{-4}	1.7×10^{-4}	5.8×10^{-4}	1.3×10^{-3}		8.3×10^{-3}	3.5×10^{-3}	1.5×10^{-2}	8.7×10^{-2}
	Cu^{++}	9.2×10^{-4}	2.2×10^{-3}	2.5×10^{-3}	7.0×10^{-3}	1.1×10^{-3}	4.0×10^{-3}	1.4×10^{-2}	4.0×10^{-2}	1.9×10^{-1}
	Zn^{++}	1.0×10^{-4}	2.8×10^{-4}	4.0×10^{-4}	1.0×10^{-3}	1.4×10^{-3}	5.2×10^{-4}	1.2×10^{-3}	1.2×10^{-2}	7.2×10^{-2}
	Co^{++}	2.6×10^{-5}	2.0×10^{-4}	7.0×10^{-5}	$<10^{-5}$					
	Ni^{++}	1.7×10^{-5}	1.5×10^{-4}	$<10^{-5}$	$<10^{-5}$					
	K^{+}	4.9×10^{-5}	2.4×10^{-4}	2.0×10^{-4}	5.0×10^{-4}					
	Hg^{++}						6.2×10^{-1}	6.1	33.4	12.8
	Fe^{+++}						3.8×10^{-2}	2.8×10^{-1}	5.4	30
Orion 94-82 A	Cd^{++}							4.8×10^{-1}		
	Zn^{++}							3.8×10^{-3}		
	Ca^{++}							6.7×10^{-4}		
	Ni^{++}							4.2×10^{-1}		
	Fe^{+++}							1.8		
	Cu^{++}, Hg^{++}, Ag^{+} must be $<10^{-7}$M at all times									
Orion 92-82 (ref. 30)	Cu^{++}	2.6	1.4×10^{-2}	1.8×10^{-2}						
	Ca^{++}	3.5×10^{-2}	3.4×10^{-1}	2.6						
	Ni^{++}	1.1×10^{-2}	2.8×10^{-1}	1.3						
	Zn^{++}	3.0×10^{-2}	2.4×10^{-1}	3.0						
	K^{+}	3.0	$3.0\times10^{+2}$	$3.5\times10^{+4}$						
		10^{-2}	10^{-3}	10^{-4}						

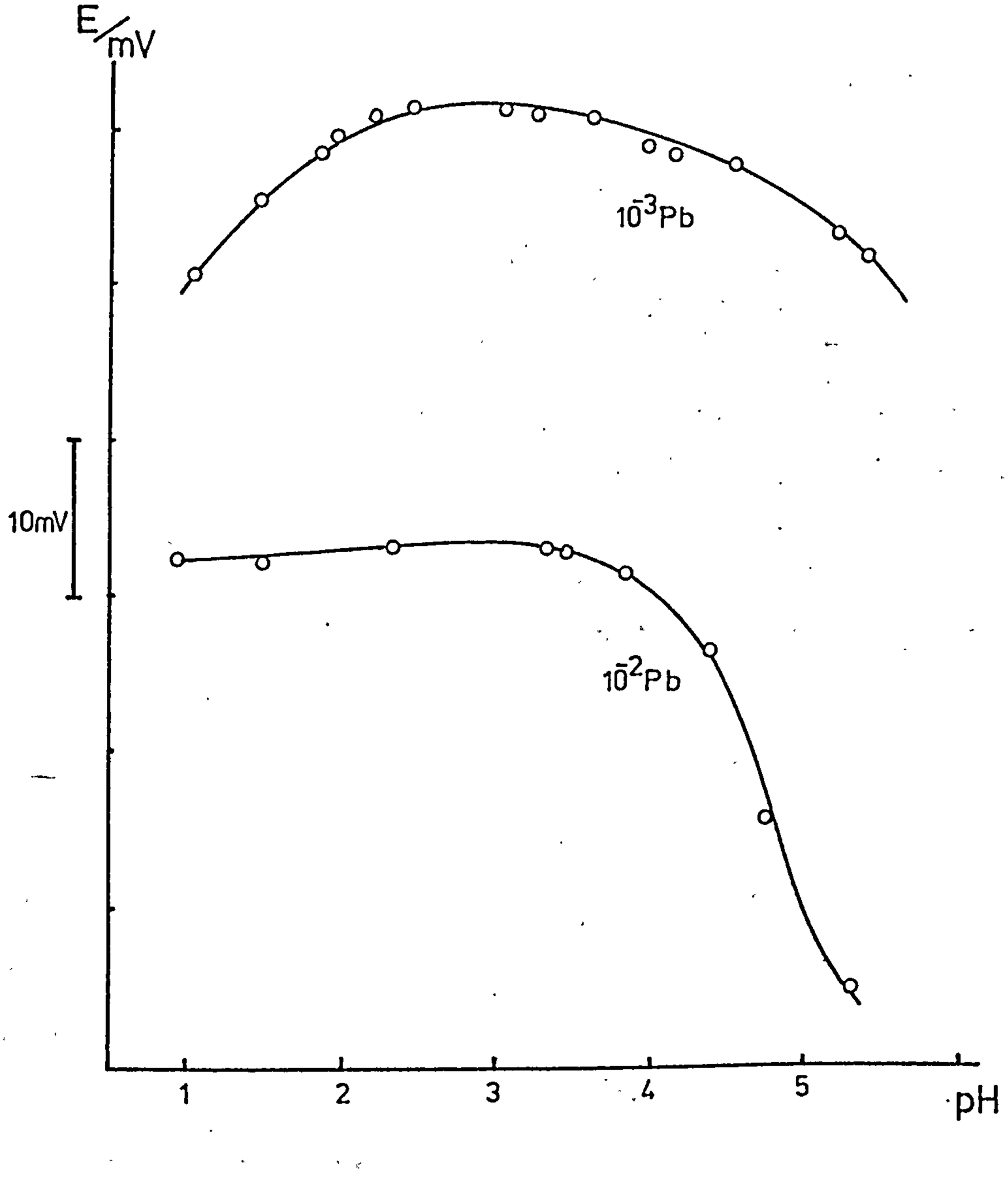
electrode were introduced to solutions of lead nitrate. The pH of the solution was adjusted by addition of concentrated nitric acid and sodium hydroxide while the pH and potential of the lead electrode were read and recorded by independent high impedance meters. The results of the changes in potential with pH, plotted in Figure 12.4.VI, show that the electrode does not respond to any great extent to pH in the range 1.5 to 4. At low pH ($\ll 1$) the membrane shows response to a hydrogen ion interference and at high pH, just before a precipitate of $Pb(OH)_2$ is visible, the membrane potential becomes more positive, usually indicative of an increase in lead concentration. This is possibly due to removal of hydrogen ions from the virginiamycin, allowing an increase in the level of lead saturation of the membrane. For the best reproducibility the pH should be kept in the range 2.5 to 3.5 for lead concentrations of 10^{-3} to 10^{-2} molar. A supernernstian response may be obtained if pH values outside of this range are used.

12.4.6. Time Response

Initial studies were made on the time response of the virginiamycin electrode. The response of the membrane when removed from one solution and placed in another, is known as the 'static' response time. The membrane responds reasonably quickly, 5 to 20 seconds being needed to reach equilibrium. The 'dynamic' time response (response to a concentration change in solution) was also studied. 10^{-1}M lead nitrate solution was injected into a 10^{-3}M solution

Figure 12.4—VI

pH Response of the Lead Electrode



(in a ratio 1:10) (ionically buffered to 10^{-1}M) to raise the lead concentration to 10^{-2}M and the electrode potential change was recorded. The response of this membrane in pure solutions is similar to other immobilized membrane electrodes (66), showing a very rapid initial response to a concentration change, followed by a period of much slower potential change, probably due to site mobility and exchange reaction kinetics within the membrane. A typical response profile to a decade concentration change is shown in Figure 12.4.VII. The value of t_{95}, the time taken to reach 95% of the potential change, being 4.9 seconds. This is a sufficiently fast response to enable the membrane to be used as the sensor in automated measurements, and as the endpoint detector in titrations.

12.5. The Effect Of Variations In the Antibiotic Membrane

12.5.1. Solvent Changes

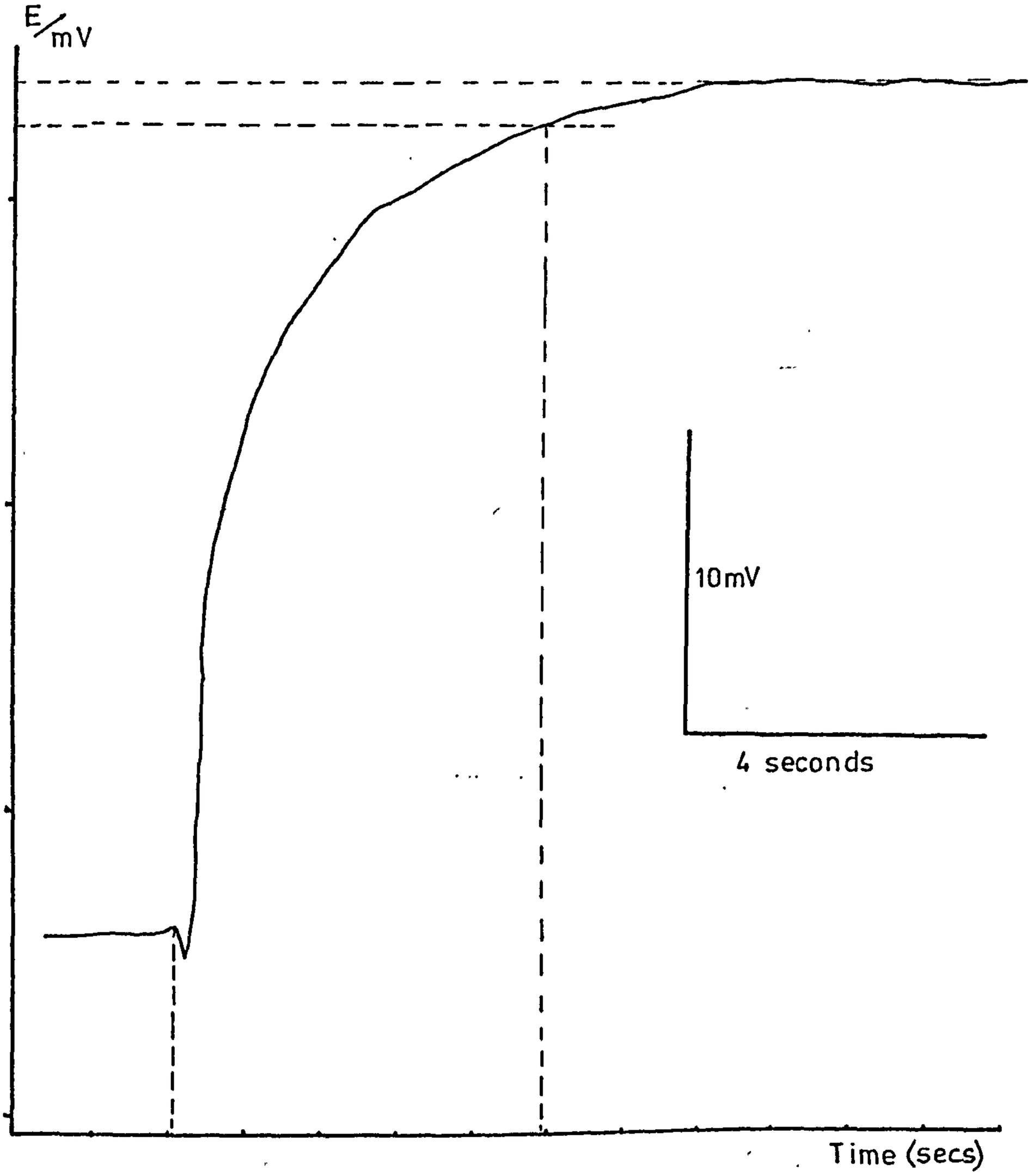
Although dioctyl adipate was used as a plasticising solvent in the lead electrode, phosphates are frequently used, e.g. in the calcium electrode. Also the selectivity of the calcium electrode has been shown to increase as the molecular weight of the solvent is increased (109). To compare this effect with that of the lead electrode a high molecular weight phosphate was used as a solvent for the antibiotic virginiamycin (M).

The membrane was prepared as previously described, using tri-n-decyl orthophosphate as the plasticising solvent. When the membrane was calibrated in lead solutions the slope of the calibration graph decreased to 20 mV/decade activity

Figure 12.4.-VII

Time Response of the Lead Electrode

($10^{-3} \rightarrow 10^{-2}$ M)



and the potential was much less reproducible (± 5mV on repeated calibrations of the same solution). A determination of the selectivity over calcium by method Ia proved to be very concentration dependant due to the non linear response of the membrane. The selectivities, given in Table 12.5.-I, show that the membrane is more selective towards calcium at higher concentrations, and is therefore unsuitable as a lead electrode.

Table 12.5.-I Variation In Selectivity Over Calcium Of The Virginiamycin (M)/Tri-n-decylorthophosphate Lead Electrode

$Conc^n (pM^{2+})$	1	2	3	4
$K^{pot}_{Pb.Ca}$	14.4	1.99	1.01	0.20

12.5.2. Antibiotic Change

Virginiamycin factor 'S' was dissolved in the same solvent as the 'lead' membrane using bis-ethylhexyl adipate as the plasticiser. Factor 'S' was found to be much more soluble in this solvent, but the concentration was kept constant for comparison.

Initial tests on permselectivity showed that the membrane responded to cations and not anions, although the sensitivity towards monovalent ions was much higher than had previously been encountered. This observation was corroborated when selectivities in pure solutions were calculated by method Ia for comparison (Table 12.5.-II).

Table 12.5.-II Selectivities Of Virginiamycin (S) Membrane At 10^{-1}M By Method Ia

Ion (J)	Ca	Cd	Cu	Co	Zn	Ni	Na
$K^{pot}_{Pb(J)}$	3.3×10^{-5}	8.6×10^{-5}	6.1×10^{-5}	2.5×10^{-7}	1.7×10^{-6}	1.1×10^{-7}	0.11
Ion (J)	K						
$K^{pot}_{Pb(J)}$	1.99						

The virginiamycin (S) membrane could be used as a very good lead selective electrode if the response towards monovalent ions could be reduced, i.e. by using a solvent with a higher dielectric constant.

12.6. Application of the Virginiamycin (M) Lead Electrode

In order to obtain accurate results in the analysis of lead solutions and to enable the measurement of various anions, the lead electrode was used as the endpoint sensor in potentiometric titrations.

12.6.1. Lead/Sulphate Titration

One of the most useful applications of the lead electrode is its use as a sensor in the titration of the sulphate anion. To evaluate the Virginiamycin membrane in this respect lead nitrate was titrated with nickel sulphate ($Pb^{2+} + SO_4^{2-} \longrightarrow PbSO_4$).

Lead nitrate (5 mls, 10^{-1}M) was diluted and ionically buffered with potassium nitrate (50 mls, 10^{-1}M). Nickel sulphate (10^{-1}M) was added from a burette with constant stirring. The electrode potential was measured with a voltmeter with respect to a saturated calomel electrode and a saturated potassium nitrate bridge.

A typical titration curve, shown in Figure 12.6.-I, gives an easily defined endpoint which is not adversely affected by the presence of the nickel ion.

12.6.2. Lead/Dichromate Titration

A titration system which is almost as common as the previous example, is the precipitation of lead by dichromate. To evaluate the membrane as an endpoint sensor, lead nitrate was titrated with potassium dichromate.

$$2\ Pb^{2+} + Cr_2O_7^{2-} + H_2O \rightarrow 2PbCrO_4 + 2H^+$$

Lead nitrate (5 mls, 10^{-1}M) was diluted with acetate buffer (pH 6.2, 50 mls) and titrated with potassium dichromate (10^{-1}M) with constant stirring. The same experimental arrangement was used as for the previous titration.

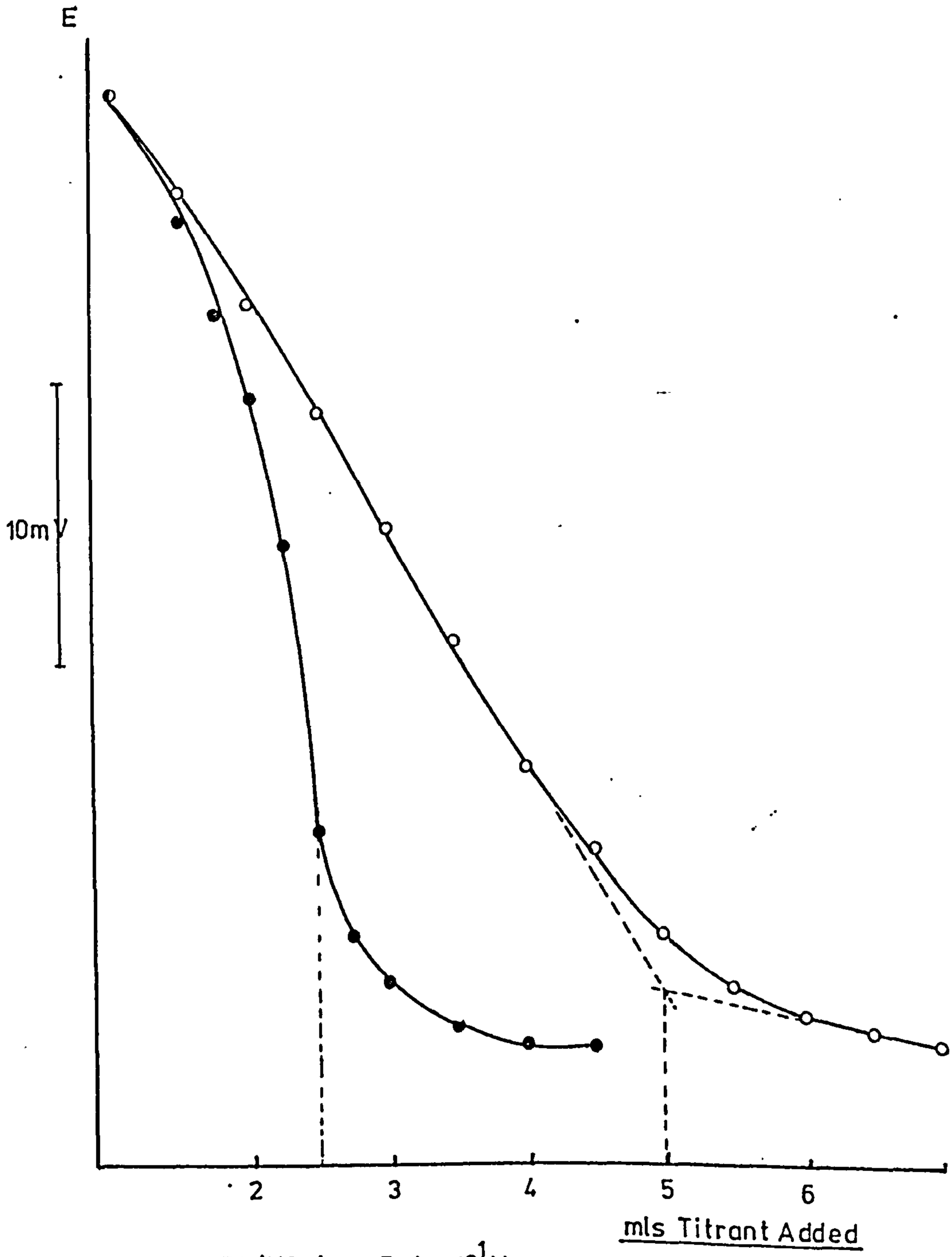
The titration curve (Figure 12.6.-II) is not an 'L' shaped plot as for the sulphate titration but is an 'S' shaped plot with a definite inflection point, possibly due to the change in response of the membrane towards concentration at this pH. The use of the dichromate caused the membrane to become yellow in colour, but neither the electrode's sensitivity nor selectivity was impaired by this effect.

12.6.3. Lead/E.D.T.A. Titration

Lead can be titrated complexometrically with E.D.T.A. to give a colourless, soluble product. Lead nitrate (10 mls, 10^{-3}M) was diluted to 30 mls with pH to buffer (NH_3/NH_4Cl) after the addition of tartaric acid to prevent precipitation of lead hydroxide. Solochrome Black T was added as a visual indicator. E.D.T.A. (10^{-3}M) was added from a burette, stirring continuously.

Figure 12.6.-I,II

Precipitation Titrations to a Lead Electrode Endpoint

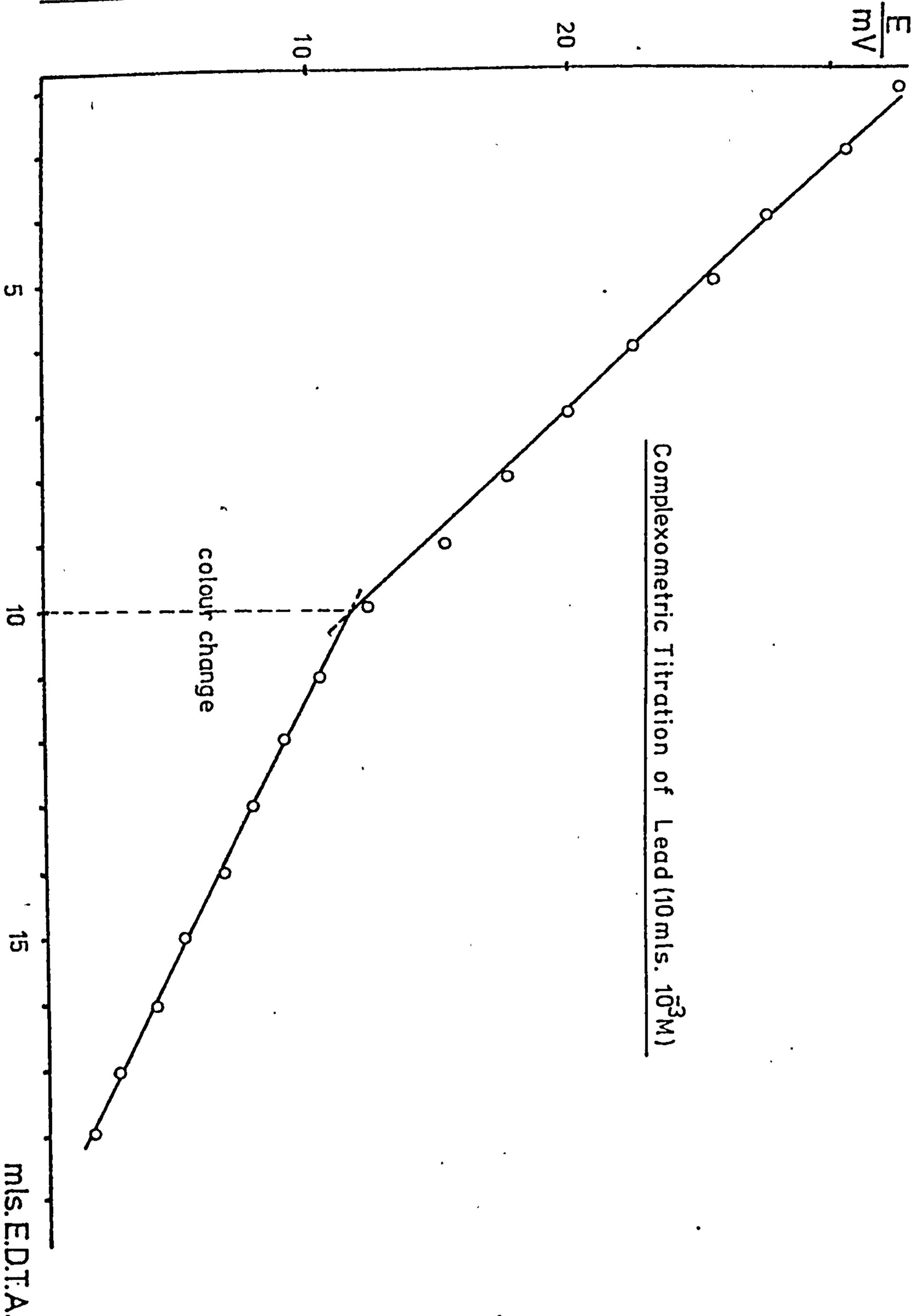


$Pb(NO_3)_2$, 5mls. 10^{1} M

I o $NiSO_4$ 10^{1} M

II • $K_2Cr_2O_7$ 10^{1} M

Figure 12.6.-III



The endpoint of the titration (Figure 12.5.-III) was easily found and corresponded to the colour change of the indicator. The complexometric titration has the advantage that no precipitates are formed during the reaction, which can be very important when the titration is automated,as precipitates tend to accumulate and block pump tubing. The E.D.T.A. titration has only a small change in gradient at the endpoint however, and this may prove a disadvantage in automation by gradient titration techniques with a virginiamycin electrode as endpoint sensor.

12.7. Conclusion

The virginiamycin antibiotic provides an ion exchanger that is very selective towards lead. The incorporation of the exchanger into a P.V.C. membrane provides an ion selective electrode which is superior in terms of sensitivity and selectivity to other electrodes available commercially. The electrode has been applied as an endpoint sensor in various titration systems to give an easy and accurate method of analysis for lead, sulphate, dichromate and other complexing species, in the presence of many heavy metals such as copper, cobalt and nickel without interference, and promises to become a valuable analytical tool.

CHAPTER 13

The Cyanide Responsive Silver Iodide Ion Selective Electrode. Application To The Measurement Of Cyanide In The Presence Of Sulphide

13.1. Experimental

13.2. Results and Discussion

13.2.1. Evaluation of the Membrane

13.2.2. Measurement of Cyanide in the Presence of Sulphide by Direct Potentiometry

13.2.3. Measurement of Cyanide in the Presence of Sulphide by Potentiometric Titration with Silver

13.2.4. Measurement of Cyanide in the Presence of Sulhpide by Potentiometric Titration with Nickel

13.3. Conclusions

The cyanide electrode is based on a silver iodide precipitate compressed into a pellet in a matrix of silver sulphide. The membrane responds to cyanide via the reaction:

$$AgI + 2CN^- \longrightarrow Ag(CN)_2^- + I^- \qquad 13.1.$$

The sulphide can be considered as a chemically inert substance through which silver ions, responsible for charge transfer, are able to move (26). Any ions which are able to complex with,precipitate or release silver ions will therefore constitute an interference. The principal interferents of the cyanide membrane are silver ions themselves, iodide and sulphide; the selectivities towards chloride, bromide and sulphate ($K^{pot}_{CN^-/X^-}$) are 10^{-5}, 10^{-3} and 10^{-5} respectively.

A model for the response of the cyanide selective membrane electrode has been described,based on a diffusion layer existing between the membrane and the bulk of solution. This layer is formed by the rapid removal of cyanide ion according to equation 13.-1 at the membrane surface (110). With this model a mean selectivity ratio of $K^{pot}_{I^-/CN^-} = 1.6$ was obtained, indicating that only small changes in iodide activity could be tolerated while monitoring cyanide if large errors were to be avoided. Although the cyanide electrode is usually used in solutions of high pH,to prevent the formation and loss of HCN, a theory developed by Mascini (111) explains the pH response of the membrane in terms of equation 13.-2, indicating a similar response from CN^- and HCN.

$$AgI + 2HCN \overset{pH \geq 7}{\rightleftharpoons} Ag(CN)_2^- + 2H^+ + I^- \qquad 13.2.$$

The cyanide electrode can be used for the direct potentiometric measurement of cyanide activity and has been applied in many situations including the analysis of cyanide in plant materials, and fruit brandies (3,5,6,112) and in some cases the results were found to be more accurate than argentometric titration to a colour indicator.

The sulphide ion has already been described as a serious interferent for the cyanide electrode and unfortunately sulphide is frequently found with cyanide, severely limiting the number of applications of the ion selective electrode (74,113). Sulphide can be removed by precipitation with cadmium (II), Pungor having reported (114) that cadmium ions do not influence the electrode response as the cadium cyanide complex is weaker than $Ag(CN)_2^-$ and disproportionates

at the electrode surface. Wronski (115) has also shown that a hydroxy mercuribenzoic acid can be used to complex sulphide in solution.

In this work these methods of masking are studied, in conjunction with potentiometric titrations, to evaluate the use of ion selective electrodes for the measurement of cyanide in the presence of sulphide.

13.1. Experimental

The effects of different membrane compositions on the stability of the cyanide electrode were investigated by preparing two electrodes of silver iodide and silver sulphide in the ratio 95:5 and 1:1 respectively. The electrodes were placed in solutions containing potassium iodide (10^{-3}M), sodium sulphide (10^{-3}M) and potassium hydroxide (10^{-1}M), and the potentials were recorded with respect to both saturated calomel and mercuric oxide reference electrodes. A membrane was then calibrated in solutions of cyanide ion, prepared by serial dilution, which were kept at a constant ionic strength and pH with potassium hydroxide (10^{-1}M, $\gamma_{CN^-} = 0.76$).

The effects of sulphide on the measurement of cyanide by direct potentiometry were evaluated by two different techniques; by calibration and standard additions.

In the calibration technique, six solutions of cyanide, with concentrations ranging from 2×10^{-4} to 12×10^{-4}M were made up, and the potentials generated by the cyanide electrode and mercuric oxide reference electrode were measured in each. Similar measurements were made in solutions

containing added sulphide and sulphide plus masking agent.

The standard addition technique consisted of measuring the electrode potential in a solution of cyanide (10 mls, 10^{-3}M, $4x10^{-1}$M in KOH) while adding 5 μl aliquots of 10^{-2} molar sodium sulphide and repeating the procedure with either cadmium chloride or p-hydroxymercuribenzoic acid present ($2x10^{-4}$M) as a masking agent.

The effect of sulphide on the analysis of cyanide by potentiometric titration with silver to a cyanide electrode endpoint was also evaluated. Samples of cyanide (5 mls, 10^{-2}M, 10^{-1}M in KOH) alone or with added sulphide and/or cadmium were titrated with ~10^{-2}M silver nitrate. The experiment was repeated using nickel as the titrant instead of silver with an ammonia, ammonium chloride (0.5M) buffer.

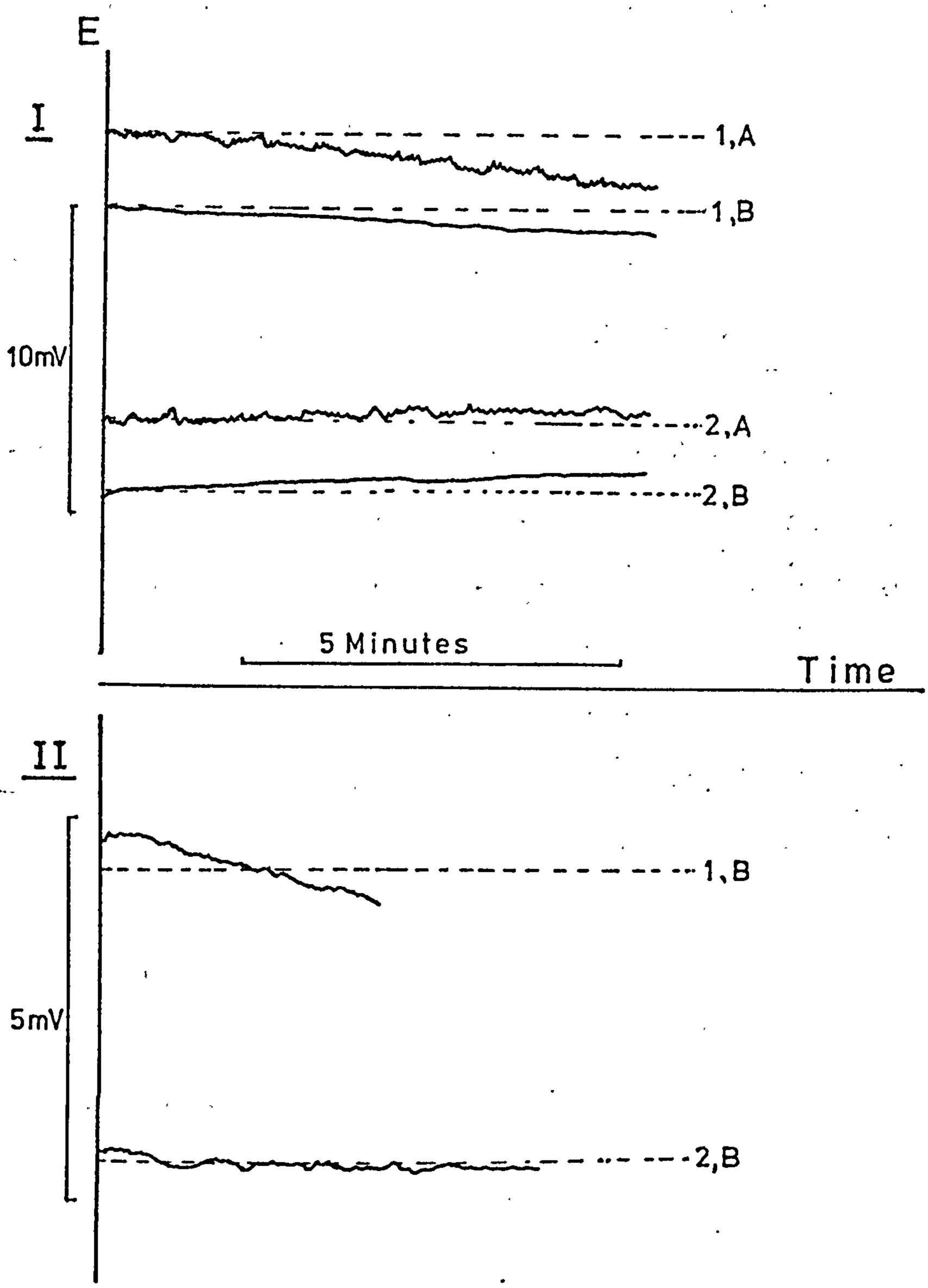
13.2. Results and Discussion

13.2.1. Evaluation of the Membrane

The initial studies on membrane composition and effects of reference electrode showed (Figure 13.2.-I) that the membrane containing 50% sulphide was more stable and that in a solution of such high pH, the mercuric oxide/KOH reference electrode exhibited less noise than the calomel electrode. A more sensitive plot of the electrode response (Figure 13.2.-II) showed that the increased sulphide concentration in the membrane matrix greatly reduces the drift in potential of the electrode in mixed analyte solutions. This stabilization can be attributed to the formation of a diffusion barrier of silver sulphide at the membrane surface.

Figure 13.2.-I/II

Evaluation of the Cyanide Membranes



A	SCE reference
B	HgO reference
1	95:5 CN membrane
2	1:1 CN membrane

When the surface of a cyanide electrode is in contact with a solution containing sulphide ion, a loose layer of silver sulphide is formed by an exchange reaction:

$$2AgI + S^{2-} \longrightarrow Ag_2S + 2I^- \qquad 13.3.$$

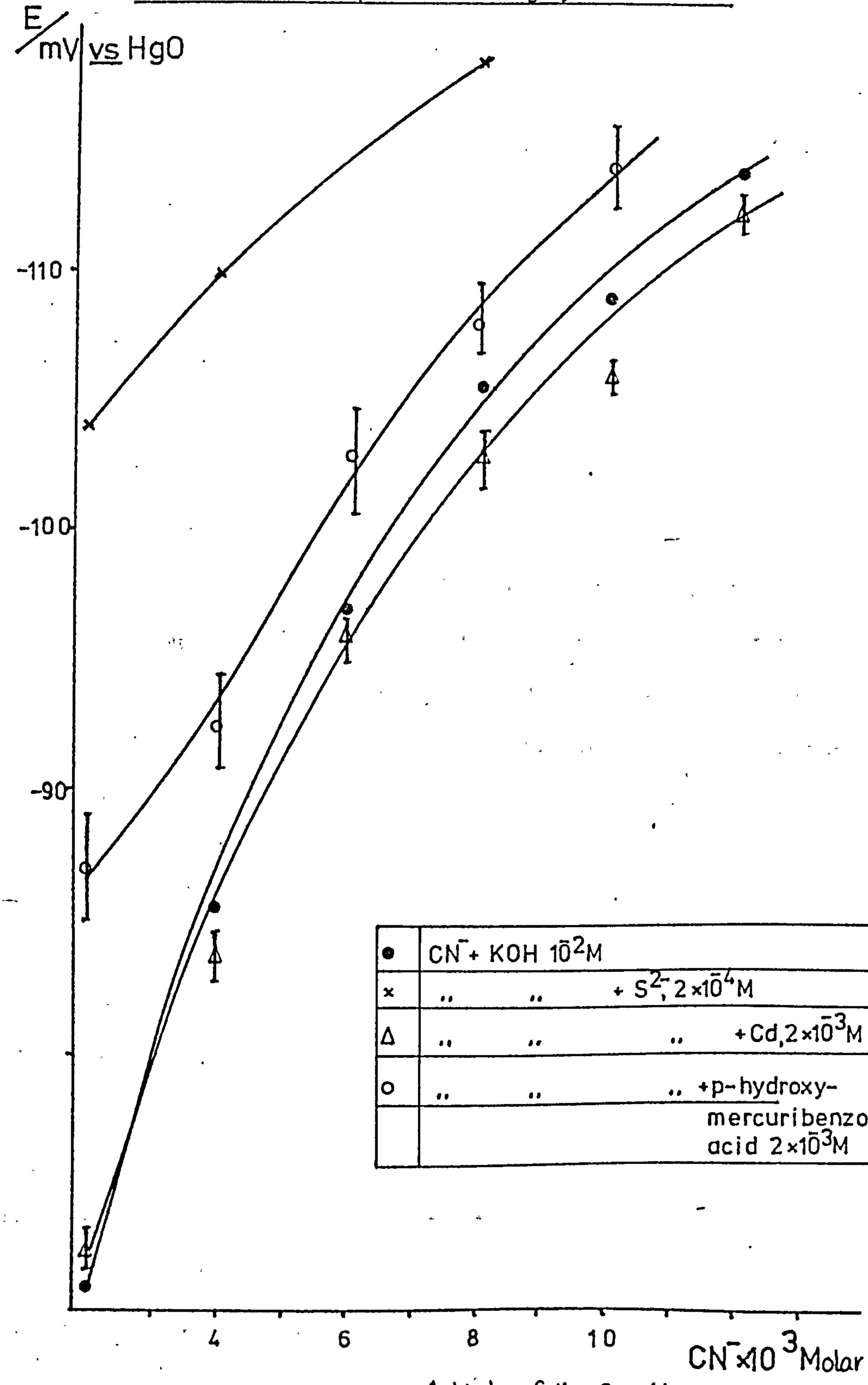
The potential generated by this ion exchange process is liable to drift as the E_o of the cyanide electrode changes, variations usually being in the order of 2-10mV per minute. The sulphide layer is also easily disturbed by stirring, and gives rise to the noise shown in the potential recordings.

It is evident from these results that the increased concentration of sulphide in the membrane mix creates an effective barrier to the formation of a loose sulphide coating, and is able to stabilize the membrane response towards cyanide in the presence of sulphide. As a ratio of iodide to sulphide 1:1 is the maximum percentage of sulphide which guarantees consistent contact between silver iodide particles; this mixture was used throughout the experiment.

Calibration of the cyanide electrode in cyanide solutions and computer analysis of the results gave a slope factor (S) of -57.3 mV per decade activity at 20°C and a value for E_o of -387.1 mV. The standard deviation of the results assuming linearity over a concentration range of 10^{-2} to 10^{-5} molar was 0.99.

Figure 13.– III

Evaluation of Sulphide Masking by Calibration



13.2.2. Measurement of Cyanide in the Presence of Sulphide By Direct Potentiometry

The results of the calibration technique are given in Figure 13.-III. Cadmium appears to be an effective masking agent but the hydroxy mercuribenzoic acid gives rise to drifting potentials and an unstable response, possibly due to the effect of mercury at the membrane.

The results for the standard addition technique (Figure 13.-IV) agree with those for the other method. Cadmium effectively masks out the interference of sulphide but the addition of p-hydroxymercuribenzoic acid, although it does change the potential of the membrane, is not beneficial. It must be pointed out that the hydroxymercuribenzoic acid used in this work is in the 'para' form and not the 'ortho' form as suggested by Wronski, however, it is suggested that this will have little effect in this application.

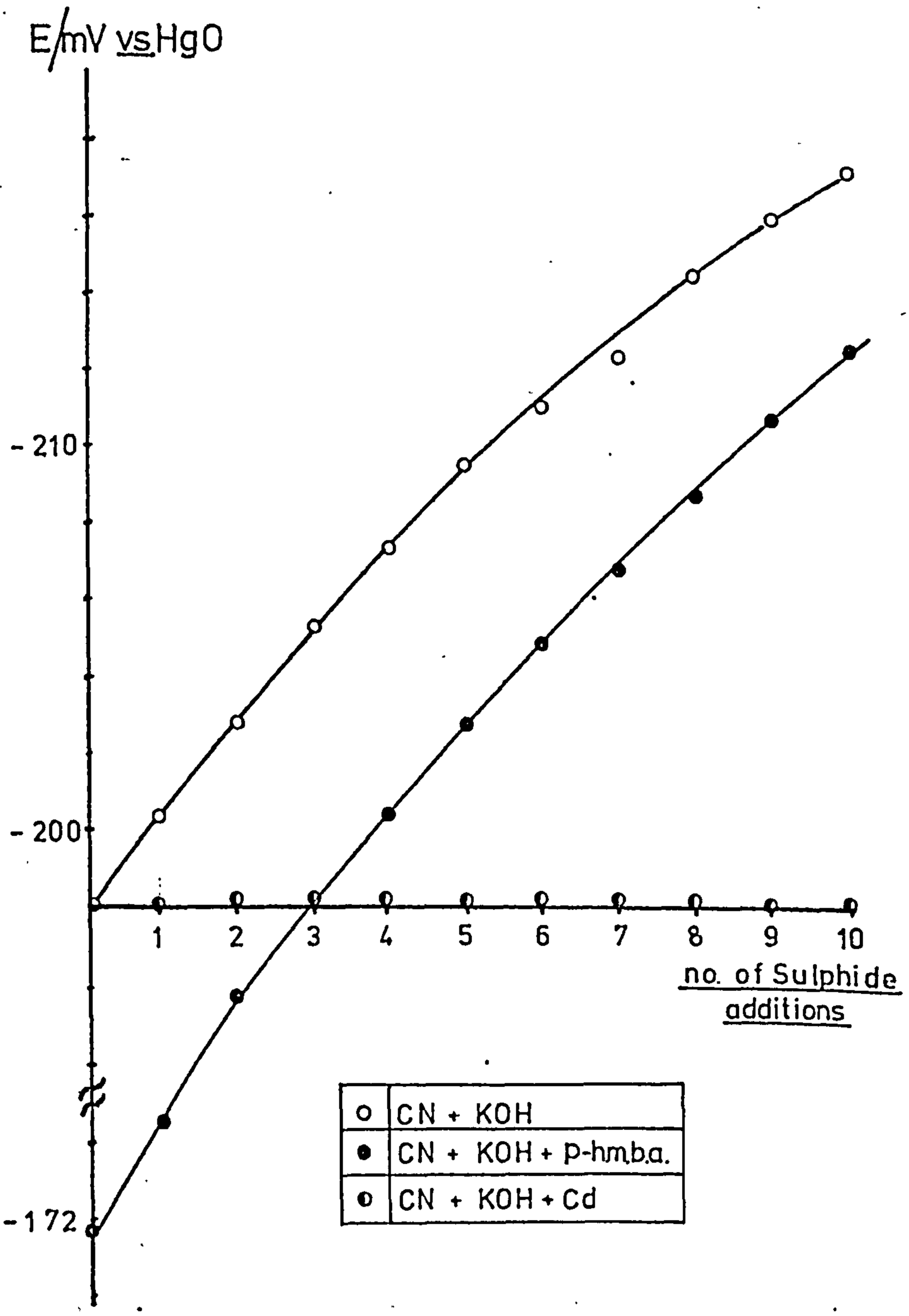
13.2.3. Measurement of Cyanide in the Presence of Sulphide by Potentiometric Titration with Silver

The results for the titration of potassium cyanide with silver nitrate are shown in Figure 13.-V. The curve for cyanide alone (13.-Va) shows an inflection point at 2.5 mls (Ag^+) due to reaction: $2\ CN^- + Ag^+ \longrightarrow Ag(CN)_2^-$ and a much more indistinct precipitation endpoint at 4.8 mls (Ag^+) due to: $Ag(CN)_2^- + Ag^+ \longrightarrow 2AgCN$.

The titration was repeated with the addition of sodium sulphide and the curve (13.-Vb) shows an inflection equivalent to the titration of total cyanide plus sulphide.

Figure 13.—IV

Evaluation of Sulphide Masking by a Standard Addition Technique



The initial formation of a black precipitate followed by a white one, indicates that the reaction follows the sequence expected from stability constants:

$$S^{2-} + 2Ag^{+} \longrightarrow Ag_2S\downarrow$$

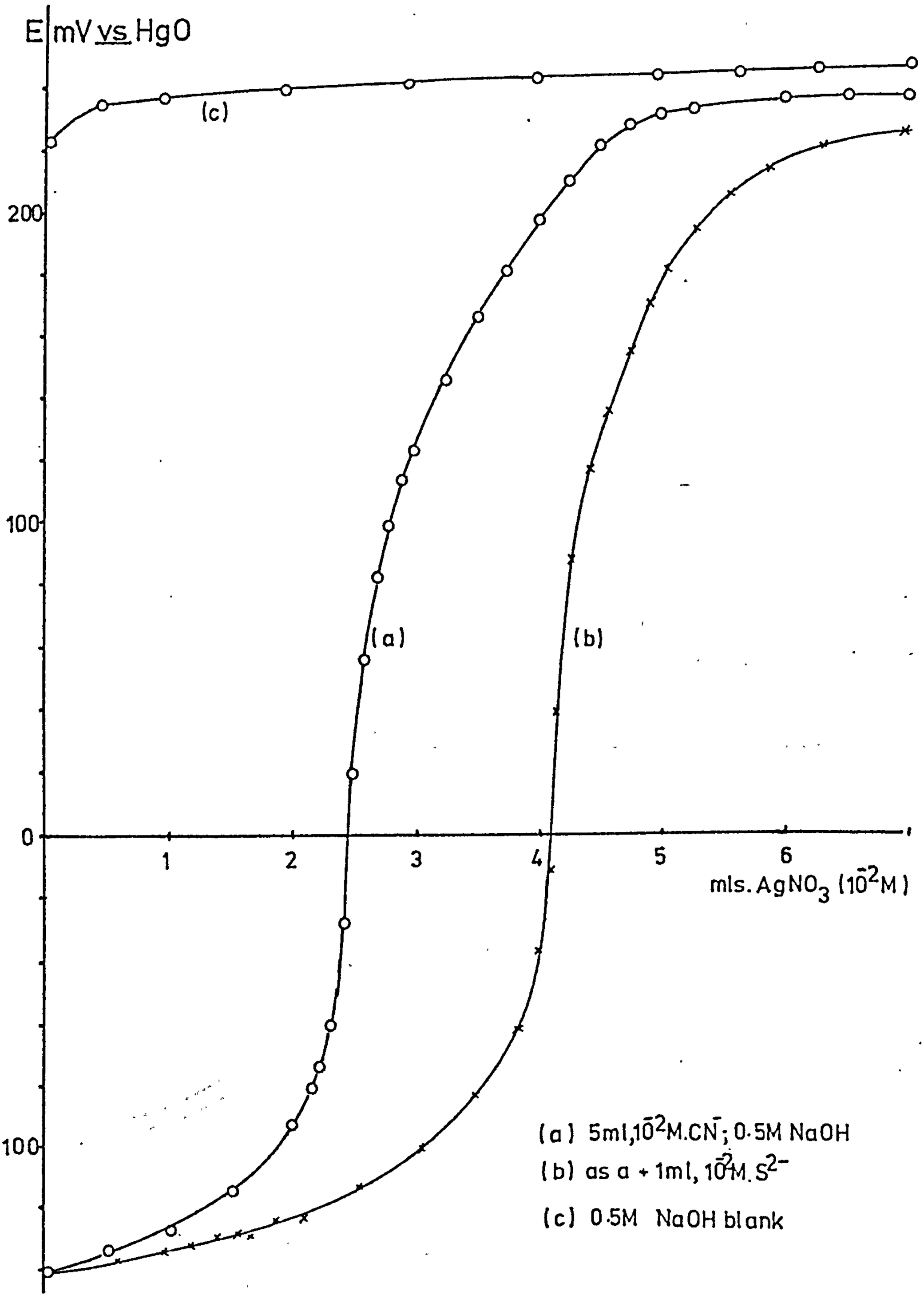
$$2CN^{-} + Ag^{+} \longrightarrow Ag(CN)_2^{-}$$

$$Ag(CN)_2^{-} + Ag^{+} \longrightarrow 2AgCN$$

Although the sulphide was titrated initially, the presence of cyanide ensured an almost constant potential and no individual equivalence points were indicated.

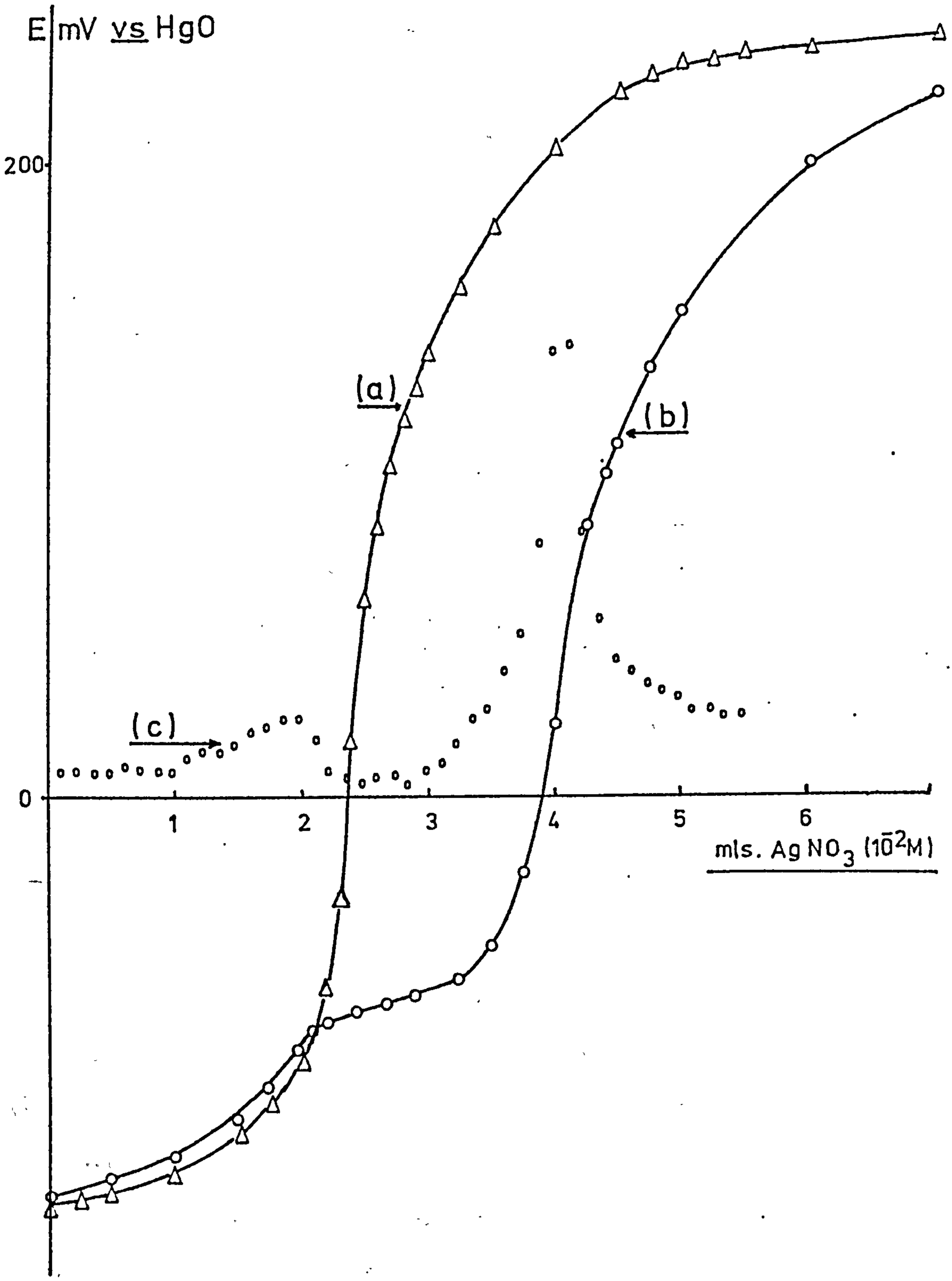
The results of cadmium masking are shown in Figure 13-VI. The addition of cadmium solution to the cyanide was accompanied by an immediate precipitation of cadmium hydroxide and the endpoint for the cyanide + cadmium titration mixture (Figure 13-VIa) occured earlier in the titration, as predicted by the theory for interferences in solution, but the difference is not large enough to cause any serious errors. The titration was repeated, adding sodium sulphide to the cadmium, cyanide mixture, (Figure 13-VIb). The two endpoints indicated at 2 mls and ~4 mls, shown clearly in the differential treatment of the results, correspond to the titration of sulphide, and sulphide plus cyanide. Cadmium can therefore not be used as a masking agent for sulphide when titrating cyanide with silver, and the reaction, $2Ag^{+} + CdS \rightarrow Ag_2S + Cd^{2+}$ is expected from inspection of the solubility products. If cadmium is to be used as a masking agent a titrant with a much more soluble sulphide, such as nickel, must be used.

Figure 13.–V



Titration of Cyanide with Silver in the Presence of Sulphide

Figure 13.-VI



(a) 5ml, 10^{-2}M. CN^-; 0·5M. NaOH + 1ml, 10^{-1}M. $Cd(NO_3)_2$

(b) as a + 1ml, 10^{-2}M. Na_2S

(c) Differential treatment of results for b

Titration of Cyanide with Silver in the Presence of Sulphide and Cadmium

13.2.4. Measurement Of Cyanide In The Presence Of Sulphide By Potentiometric Titration With Nickel

Wronski's original procedure for the independent titration of cyanide and sulphide involved the masking of sulphide and titration of cyanide with nickel to a colourimetric endpoint. The equation for the reaction is:

$$4CN^- + Ni^{2+} \rightleftharpoons Ni(CN)_4^{2-}$$

$$Ni(CN)_4^{2-} + Ni^{2+} \longrightarrow 2Ni(CN)_2 \downarrow$$

In the presence of an ammonia, ammonium chloride buffer (0.5M), nickel does not form a sulphide precipitate and will thus not titrate any sulphide present in a cyanide sample. The absence of nickel sulphide formation in this medium is explained in the following equilibria (116).

$$Ni^{2+} + NH_4HS + NH_3 \rightleftharpoons NiS + 2NH_4^+$$

$$NiCl_2 + NH_3 + H_2O \rightleftharpoons Ni(OH)Cl + NH_4Cl$$

$$Ni(OH)Cl + 7NH_3 + H_2O \rightleftharpoons Ni(NH_3)_6 \quad (OH)_2 + NH_4Cl$$

Nickel sulphate solution was added to cyanide solution with an ammonia, ammonium chloride buffer. In the titration only one endpoint was visible when the cyanide activity was monitored by a cyanide electrode and mercury oxide reference, corresponding to the formation of the $Ni(CN)_4^{2-}$ complex, a typical curve for which is shown in Figure 13-VII.

The titrations were repeated with additional cadmium solution and sulphide, the results for which are given in Table 13-VIII.

As a quantity of sulphide will require an equal amount of nickel for equivalence, it can be seen from these results that the addition of sulphide to the sample does not

have a major effect on the titration endpoint. The presence of cadmium in the sample does have an effect however and must be allowed for in any analysis of results. The titration results for a mixed sample without cadmium masking agreed well with the results for an equivalent amount of cyanide alone but in the presence of sulphide the endpoint potential change was smaller. In the case of low cyanide levels, or high sulphide levels, the potential would not change a sufficient amount for accurate analysis.

Table 13-VIII : Results of Cyanide Titration With Nickel

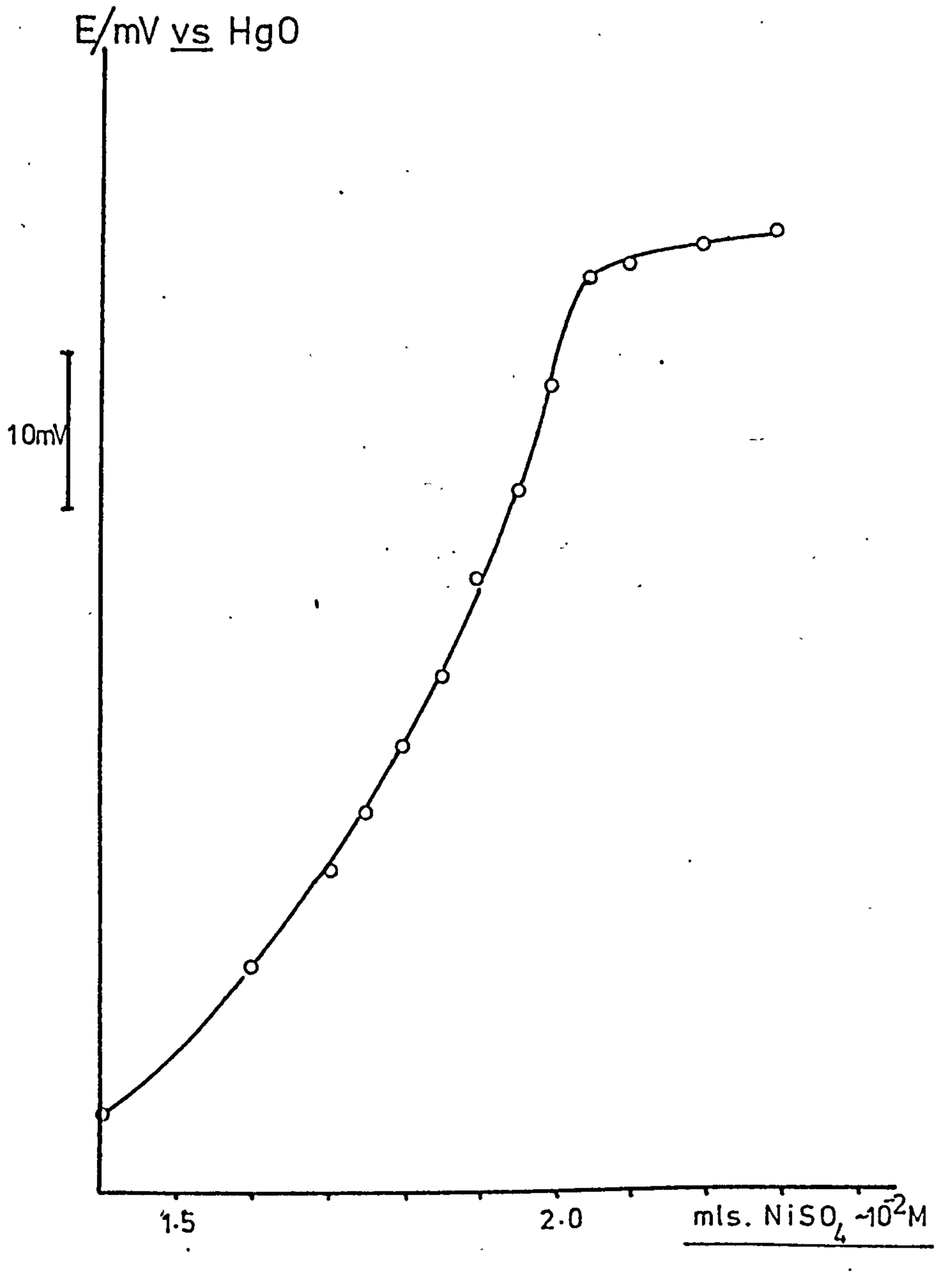
CN^- Taken mls $10^{-2}M$	S^{2-} added mls $\sim 10^{-2}M$	Cd II added mls $10^{-1}M$	Ni II vol. at endpoint $\sim 10^{-2}M \pm 0.05$	Emf Change During Titn. mV
10	0.0	0.0	1.95	89
10	0.0	1.0	2.22	80
10	0.5	0.5	2.25	70
10	0.5	1.0	2.25	72
10	1.0	1.0	2.75	72
10	1.0	0.5	2.75	83
10	1.0	0.0	1.95	60

13.3. Conclusions

Although the potential of a silver iodide membrane drifts when the electrode is placed in a solution containing sulphide ion, a diffusion barrier, provided by incorporation of 50% silver sulphide into the membrane precipitate mixture, considerably stabilizes the potential without adverse effect on the other electrode properties. When the electrode is used for direct potentiometric analysis of cyanide, cadmium ions in solution act as an efficient masking agent for sulphide whereas hydroxymercuribenzoic acid proved to be ineffective.

Figure 13.-VII

Titration of Cyanide with Nickel



10 mls, 10^{-2}M CN^- + 5 mls, 0·5M NH_3, NH_4Cl buffer

If the cyanide electrode is used as an endpoint indicator in a potentiometric titration with silver, cadmium cannot be used as a masking agent for sulphide due to the much lower solubility of silver sulphide. With cadmium in the solution, however, the sequential titration of sulphide and cyanide is apparent and individual titration values can be evaluated from the results. When a nickel titrant is used instead of silver nitrate, cadmium masking becomes unnecessary with high cyanide or low sulphide concentrations. If the cyanide concentration is low or the sulphide concentration is very high, the effect of sulphide can be removed by addition of an excess of cadmium, provided the interference of this ion (Cd) on the titration equivalence point is allowed for.

It can be concluded from this work that cyanide can be measured in the presence of sulphide using both direct potentiometric and titration analysis with ion selective electrodes in conjunction with cadmium masking and a nickel titrant.

CHAPTER 14

The Chloride Sensitive Ion-Selective Electrode: Application To The Measurement Of Chloride Concentration In Milk As A Proposed Monitor For Mastitic Infection

14.1. Experimental

14.1.1. Apparatus and Reagents

14.1.2. Analytical Procedures

14.1.2.1. Ion Selective Electrode Potentiometry

14.1.2.2. Titration - British Standard Method

14.1.2.3. Leucosite Cell Count

14.2. Results and Discussion

14.2.1. Calibration of Electrode

14.2.2. Comparison of the Potentiometric Method of Standard Additions with the British Standard Method

14.2.3. Analysis of Milk Samples

14.3. Conclusions

Infections of the udder may bring about a reduction in yield of milk and changes in its composition. The changes in composition are largely attributed to an increased leakage of body fluids into the 'healthy milk', and result in increased levels of sodium, chloride and non-casein protein and a decrease in potassium and lactose levels (117). The high levels of salts can result in an increase in electrical conductivity and a good correlation between conductivity and leucosite cell count has been reported (118). A high leucosite cell count ($> 10^6$/ml) is usually taken as a guide to infection. Measurement of milk conductivity has

been considered as a possible method of determining mastitic milk, but as yet very little practical achievement has been reported.

Chloride was proposed may years ago as an indicator of mastitic milk and a sugar-chloride coefficient, the so-called Koestler number = $\frac{100 \times Cl/\%}{\text{lactose (per cent)}}$ (119) has been extensively used. A Koestler number of from 1 to 3 is obtained from normal milk and >3.5 from abnormal or infected milk. High values may also be obtained in late lactation, however, independent of udder infection.

Changes in concentration of sodium, potassium, chloride and lactose in milk may be determined by chemical techniques, but these are usually quite complex. Electrochemical methods of analysis are attractive because of high precision and ease of automation and at the present time electrodes responsive to sodium, potassium and chloride ions are commercially available and an electrode responsive to lactate ion has also been reported. The chloride ion-selective electrode has been used to determine the chloride concentration in milk using the direct potentiometric approach (120).

In this work the chloride concentration of milk was determined by a standard addition technique and by a direct method, both of which were compared with the British Standard method of chloride analysis (121) and correlated with the leucosite cell count measured by a Coulter counter technique. The results were also correlated with a bioassay procedure to determine the bacteria present in the samples.

14.1. Experimental

14.1.1. Apparatus and Reagents

The selective electrode used in this work was an original design, developed as a combined sensing,double junction reference electrode. The reference electrode compartment is contained in an anular space surrounding the central sensing membrane and incorporates a potassium nitrate salt bridge.

The electrode potentials were measured in one of two ways:

(1) Directly (using a Corning EEL digital voltmeter)

(2) Indirectly, by a purpose-built antilog amplifier (Chapter 9)

Standard chloride solutions were prepared from dried AnalaR KCl and deionised water. Samples analysed by standard additions were diluted 1:10 with $^{M}/_{2}$ KNO_3 (AnalaR) as an ionic strength buffer.

14.1.2. Analytical Procedures

14.1.2.1. Ion Selective Electrode Potentiometry

Standard Additions: To a 25 ml sample (of whole milk diluted to 1:10 with KNO_3) sequential additions of 1 molar potassium chloride were made. The potentials were recorded directly and the results of the analysis calculated using the computer program ADDFIT (88). This program provides a computer fit for the Gran's Plot analysis of multiple additions.

Direct Potentiometry: Analysis of the milk samples by the direct method was carried out in two ways:-

(i) The electrode was dipped into whole milk and the potential was recorded after allowing 20 sec. for potential stabilization. The chloride ion activity was calculated from a calibration graph obtained by making multiple additions of chloride ion to a $^{M}/2$ KNO_3 ionic strength buffer.

(ii) In the second approach the potential was measured using the antilog amplifier. The calibration graph was prepared as in the previous method. As the chloride ion activity coefficient for whole milk medium is not accurately known all of the above methods only provide comparative procedures. Since the use of the chloride electrode is only proposed for routine screening this does not pose a serious limitation. The results obtained for the standard addition method were compared with the standard titrimetric procedure for chloride in milk.

14.1.2.2. Titration- British Standard Method 1741:1963.

The milk sample (10 ml) is pipetted into a flask, silver nitrate (10 ml,0.05N) and nitric acid (10 ml conc.) are added and the mixture is boiled. The solution is cooled, diluted and the excess silver is titrated with potassium thiocyanate (0.052N) to a faint reddish brown colour with an iron alum indicator.

14.1.2.3. Leucosite Cell Count: The results obtained in the first section were correlated with the leucosite cell count obtained by the Coulter Counter technique (122).

14.2. Results and Discussion

14.2.1. Calibration of Electrode

The chloride electrode was calibrated by placing it in a series of chloride solutions, buffered to constant ionic strength with potassium nitrate (γ = 0.755). The potential was measured with respect to a (Ag/AgCl/KCl sat // KNO_3// Sample/) reference system. The calibration graph for the electrode is shown in Figure 14.2.-I. The slope factor for the electrode response is 55 mV per decade at 19°C.

14.2.2. Comparison of the Potentiometric Method of Standard Additions with the British Standard Method

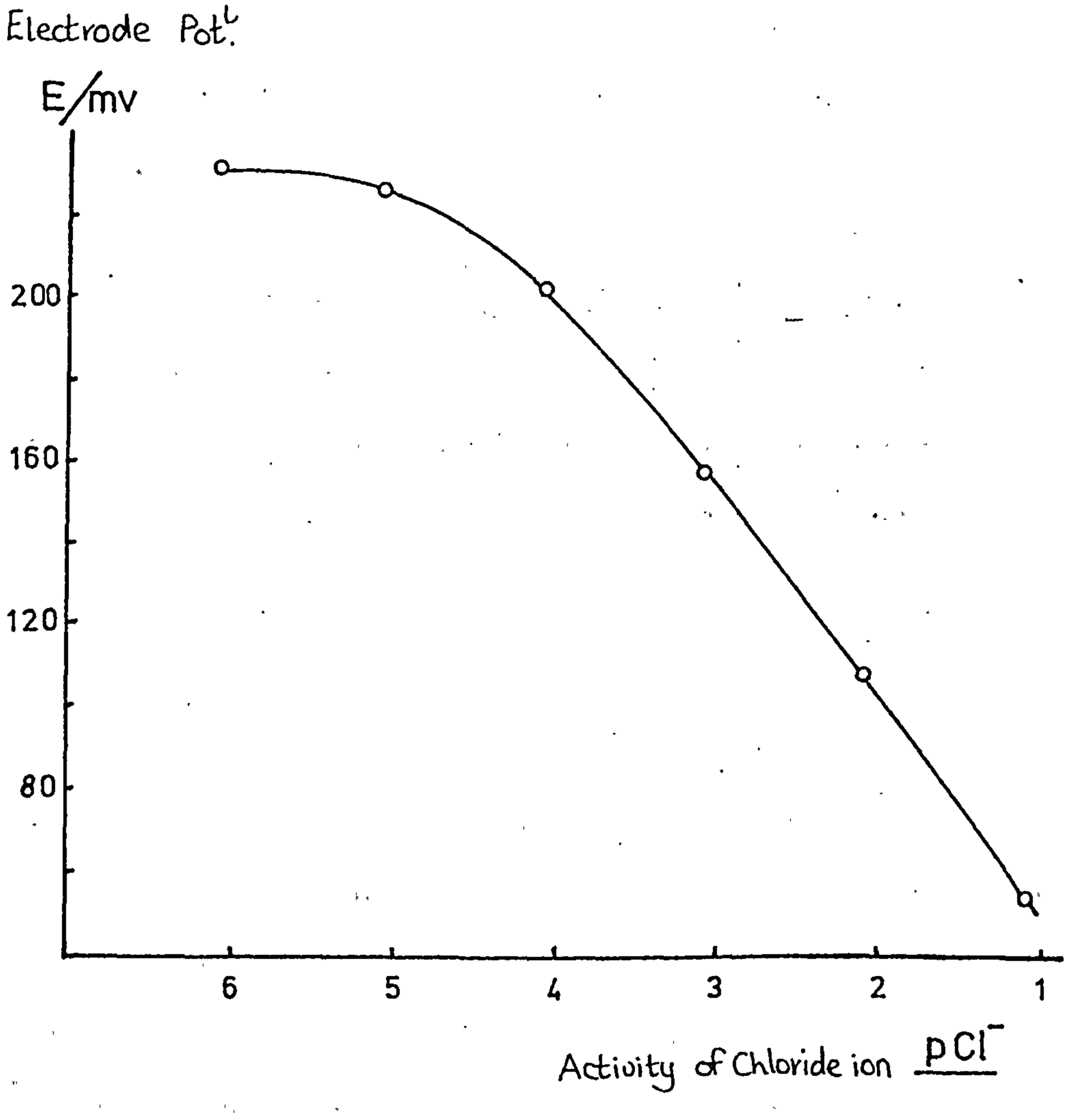
Ten replicate samples were analysed by the standard addition technique and also by the British Standard method. The results are shown in Table 14.2.-II

Table 14.2.-II

Results of Comparison of Chloride Analysis by Standard Additions and the British Standard Method

Method	Standard Addition	British Standard
Cl^- Found	2.95,3.30,3.43,3.20,3.27	3.202,3.281,3.241
x 10^2M	2.49,2.92,3.21,2.17,3.15	
Mean Cl^- (M)	3.20×10^{-2}	3.24×10^{-2}
Std. Devn. (s)	0.18	mean Devn. $(\frac{\Sigma d}{n})$, 0.03

Figure 14. 2.-I



Calibration of the Chloride Electrode

14.2.3. Analysis of Milk Samples

Milk samples, previously stabilized by addition of formaldehyde were analysed using a Coulter Counter. Two replicates of each sample were diluted 1:10 with KNO_3 ($^M/_2$) and analysed by the standard addition technique (seven 1M. Cl^- additions of 200 μl). At the same time a sample of whole, undiluted milk was retained for analysis by direct potentiometry. The results of these analyses are shown in Table 14.2.-III. Additions of chloride were made to $^M/_2$ potassium nitrate to obtain relative values for the activity of chloride found by the direct methods of analysis. The calibration graphs are shown in Figure 14.2-IV. A graphical correlation between cell count and both chloride concentration and chloride activity is given in Figures 14.2-V and 14.2.-VI.

14.3. Conclusions

The correlation between cell count and chloride concentration is very good and there can be no doubt that analysis of chloride by ion selective electrode potentiometry provides a sensitive method of determining changes in the udder. A milk sample with a chloride level in excess of 3.7×10^{-2}M for example, can reasonably be assumed to come from a cow with a bacterial disease, probably Staphylococcus aureus, unless the cow is known to be drying off.

The results for direct potentiometric analysis agree with those obtained by the standard additions method, and the use of the antilog amplifier considerably simplifies the measurement of activity by direct potentiometry.

Table 14.2.-III

Correlation of Chloride Concentration and Activity with Leucosite Cell Count

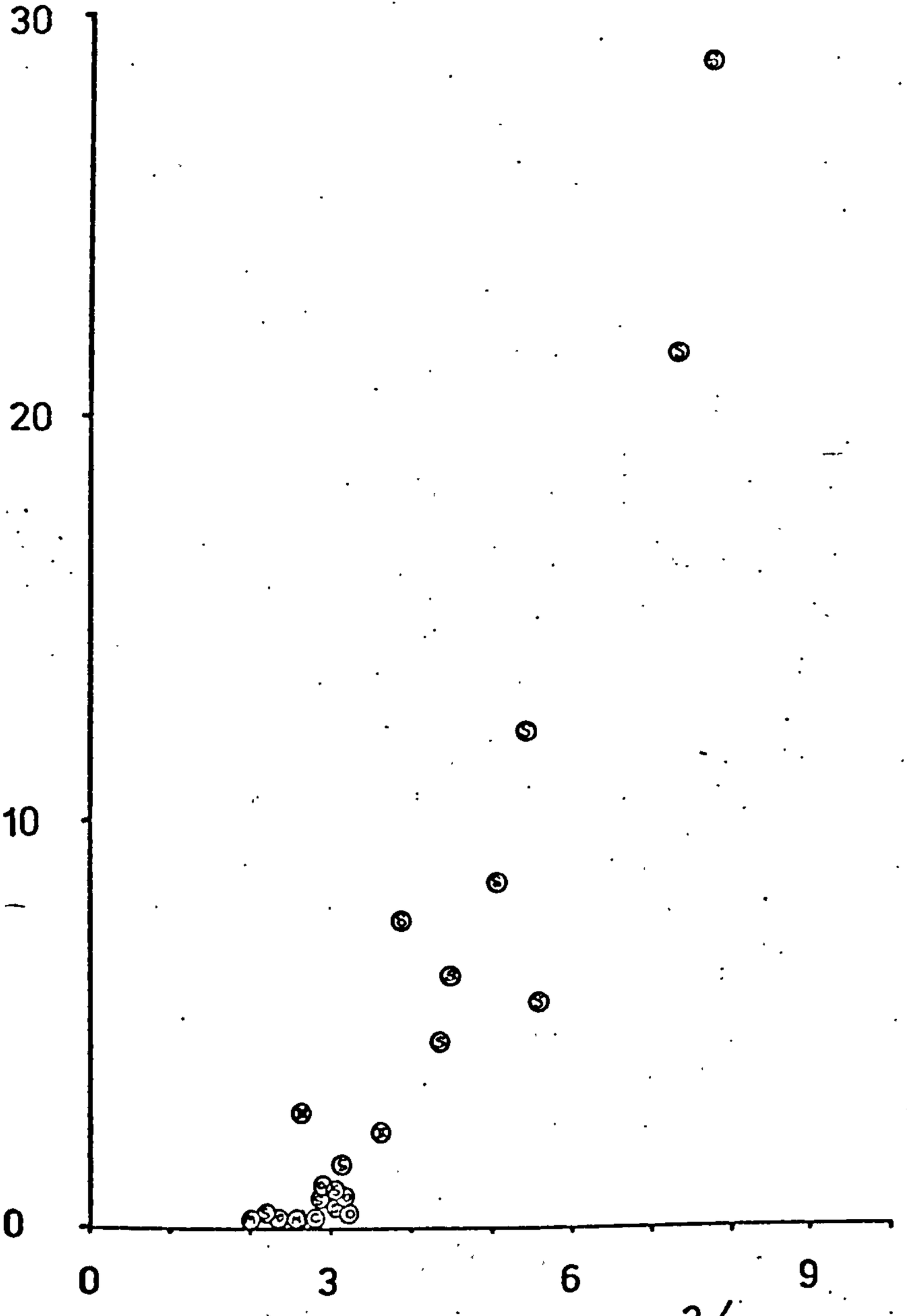
sample	Bacteria	CellCount.$\times 10^{6}$	Cl^{-} Conc^{n}.$\times 10^{2}$ (by Std. Add^{n})	rel. Cl Activity log response	rel. Cl Activ^{y}. $\log^{-1}$ response
1	D	0.630	5.98	7.8	6.35
2	S	0.890	3.12	4.3	3.70
3	III + C	0.066	2.79	3.8	3.15
4	S	0.724	2.97	4.0	4.05
5	O	0.300	3.21	4.5	3.70
6	S	4.517	4.38	5.5	4.70
7	S	0.469	2.86	4.1	3.90
8	II	2.363	3.62	4.9	4.80
9	S	0.290	2.23	3.2	2.55
10	S	12.296	5.37	7.3	6.35
11	O	0.143	2.39	3.3	2.70
12	O	1.031	2.93	4.0	3.35
13	S	7.494	3.90	6.0	4.62
14	III	0.311	3.09	4.1	3.95
15	S + D	28.933	7.63	10.6	10.15
16	S	21.685	7.25	10.0	10.35
17	D	2.063	7.06	11.0	7.85
18	S	1.796	3.17	4.5	4.05
19	III + M	0.242	2.27	2.9	2.95
20	S	8.558	5.09	7.5	7.00
21	S	5.556	5.64	8.6	6.35
22	III	2.812	2.61	3.7	3.35
23	S	6.086	4.58	5.5	5.40
24	M	0.134	2.67	3.5	2.85
25	C	0.050	2.85	4.4	3.35
26	M	0.255	2.06	3.2	2.95
27	O	0.436	3.19	3.5	3.45

Figure 14.2.-IV

Calibration Graphs of the Chloride Electrode in M/2 Potassium Nitrate

Figure 14.2.-V

Correlation of Cell Count with Chloride Concentration



S	Staphylococcus aureus
II	Streptococcus dysgalactiae
III	Streptococcus uberis
C	Cornebacterium bovis
M	Micrococci
O	No bacteria
D	Cow drying off

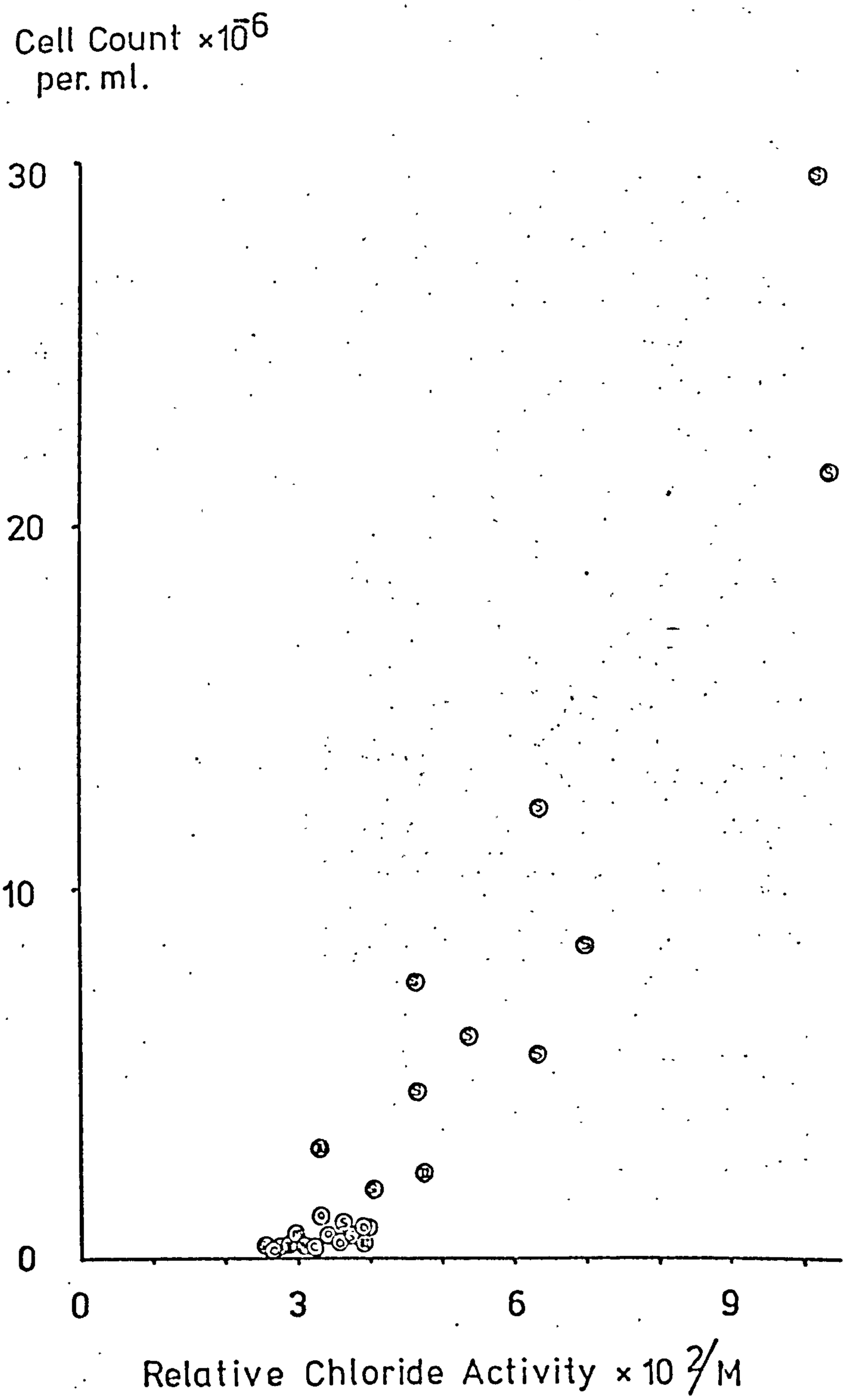


Figure 14.2.-VI

Correlation of Cell Count with Chloride Activity

The calibration graph of activity vs. potential is linear, facilitating interpretation of results and the large potential output from the device, in the order of 600 mV for an activity change of one decade (10^{-2} - 10^{-1}M), increases the accuracy possible in potential measurement, or allows the use of lower resolution (and therefore cheaper) potential measuring devices. Direct potentiometry with the antilog amplifier provides an inexpensive and rapid method of analysis that can be carried out in whole milk in individual samples or incorporated into an automated system.

Ion selective electrode analysis of chloride can therefore be used as a screening method of disease indication. Further investigation of the method to provide data on the normal levels of milk constituents, their fluctuation in absence of infection and long term variation would be of value. It is probably that the use of accurate, inexpensive and eventually automated methods of analysis using ion selective electrodes will permit small changes in levels of milk constituents to be measured, giving an indication of the occurrence of abnormal milk composition.

CHAPTER 15

The Sulphate Sensitive Ion Selective Electrode

15.1. Experimental

15.1.1. Preparation of the Membrane

15.1.2. Evaluation of the Membrane

15.2. Results and Discussion

15.3. Conclusion

Many attempts have been made to produce an electrode selective to the sulphate anion. Earlier work carried out with membranes of barium sulphate in many different forms have shown that, while it is easy to obtain a response to sulphate, it is very difficult to make an electrode with any degree of selectivity (30,31,32).

A publication by Rechnitz et al. (81) has indicated that a sulphate selective electrode may be prepared from a mixture of the sulphides of silver and lead and lead sulphate. In this study the electrode was prepared and evaluated for use as a sensor in automated sulphate analysis, by both direct and titration methods.

15.1. Experimental

15.1.1. Preparation of the Membrane

The precipitate mixture suggested by Rechnitz contained 32 mole % Ag_2S, 31 mole % PbS, 32 mole % $PbSO_4$ and 5 mole % Cu_2S. To prepare this mixture of precipitates, lead and silver nitrates in the correct proportions were dissolved in water, and H_2S from a cylinder was passed through slowly while stirring. The precipitate was filtered off and washed with water and alcohol then dried in an oven at 80^{o}C. Cuprous sulphide was

formedby hydrogen sulphide precipitation from cuprous chloride, and lead sulphate was precipitated from lead nitrate by sulphuric acid. The precipitates were weighed, and ground together in a mortar and pestle to a fine grey powder.

The sulphate mixture (0.168 grams) was placed in a 6 mm diameter die, evacuated and subjected to a load of two tons at 200°-300°C for twenty hours. After cooling, the membrane was stuck into a P.V.C. electrode end cap, with a filling solution of 10^{-3}M silver nitrate, before soaking in alternate solutions of silver nitrate/lead nitrate and sodium sulphate for two days.

15.1.2. Evaluation of the Membrane

The electrode was calibrated in solutions of sodium sulphate and then in solutions ionically buffered with sodium nitrate. The membrane was briefly tested for response to halide and lead ions and its use as a sensor in sulphate titrations was investigated.

15.2. Results and Discussions

The initial calibration of the sulphate membrane is shown in Figure 15.2.-I. The response is linear from 10^{-2} to 10^{-5} molar with a slope (S) of -33 mV (20°C). The noise level of the electrode was high however, the potential oscillating up to 3 mV either side of the mean value. Subsequent calibrations are given in Table 15.2.-II, and indicate the amount of hysteresisor memory effect found in the membrane even though the potential was allowed to stabilize for six minutes between each reading.

Figure 15.2.-I

Initial Calibration Graph of the Sulphate Electrode

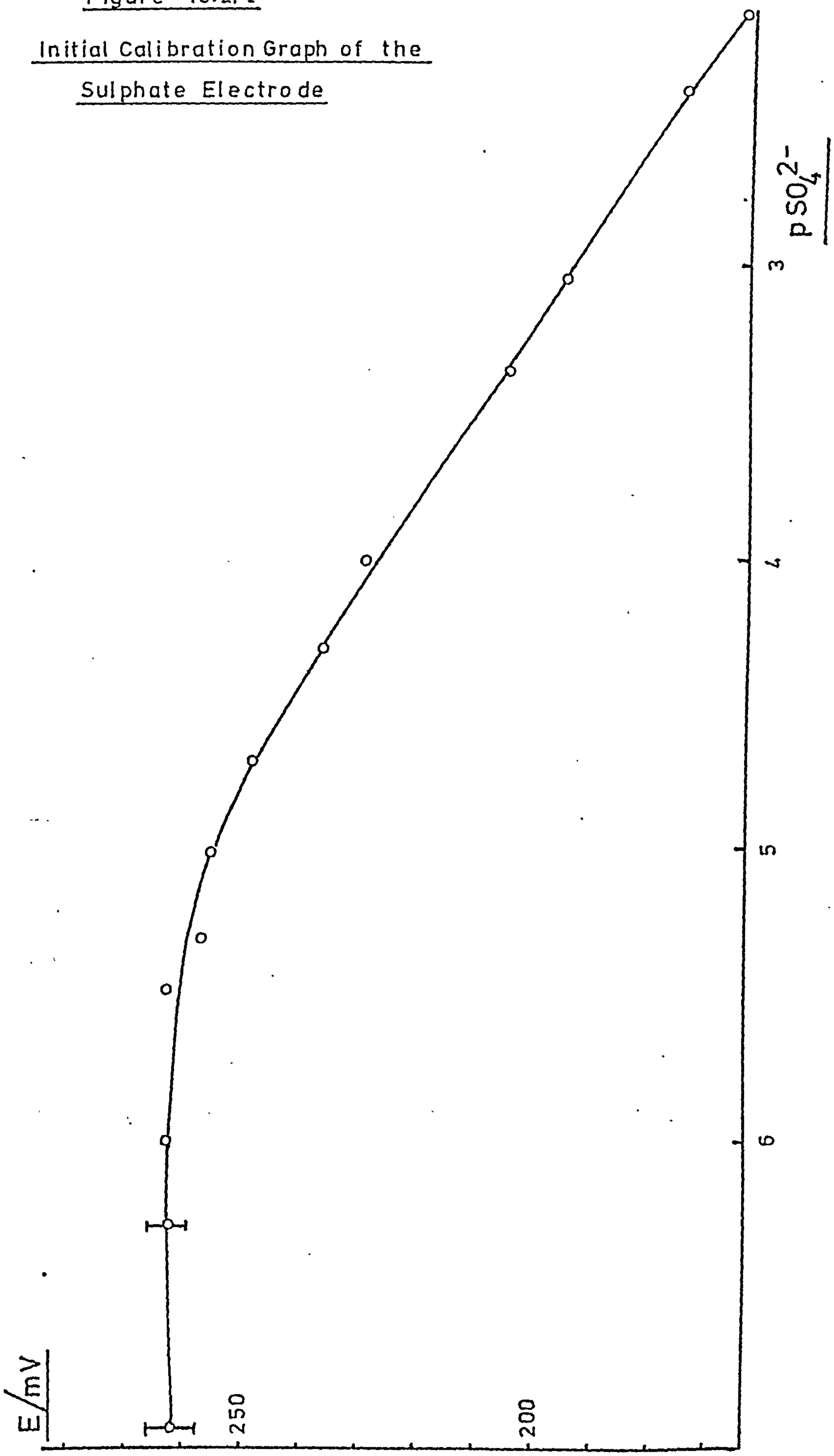


Table 15.2.-II

Calibrations of the Sulphate Membrane

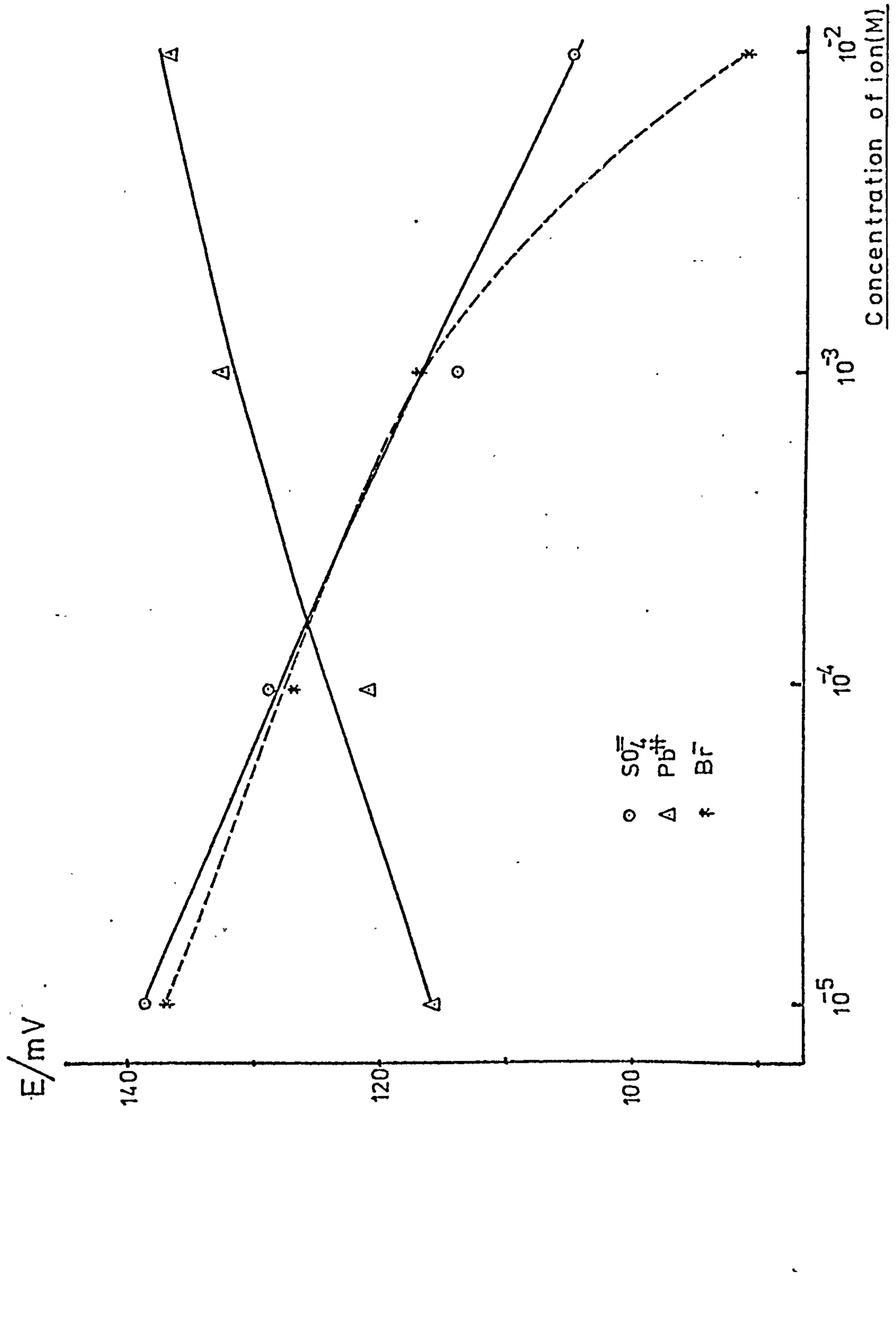
Conc. Change	Conc of Sulphate (Molar) 10^{-4}	10^{-3}	10^{-2}	10^{-1}	E vs. S.C.E. mV.
increasing	192	172	147	113	
decreasing	194	175	152	113	
increasing	194	183	165	122	
decreasing	188	171	155	122	
Mean Value	191.±2	175±5	154±5	117±4	

When an attempt was made to measure the response of the membrane to sulphate in a sodium nitrate ionic buffer (10^{-1}M), it gave a reduced response of 10 mV/decade between the concentrations 10^{-1} to 10^{-3} molar and no response below 10^{-3} molar. After soaking in metal nitrates a further twelve hours (a total life time of four days) the electrode slope reversed, and it ceased to respond to the sulphate anion. Repeated soaking and grinding of the membrane surface failed to restore the response. Several new membranes were prepared but none showed a life time in excess of five days, and the majority (6/10) failed to work at all.

The response of a membrane towards bromide and lead ions was investigated. The results shown in Figure 15.2.-III, indicate that the selectivity of the membrane is not good, although the response of the membrane to monovalent bromide is reduced to 10 mV/decade from a Nernstian value of 59.

Figure 15. 2.-III

Responses of the Sulphate Membrane



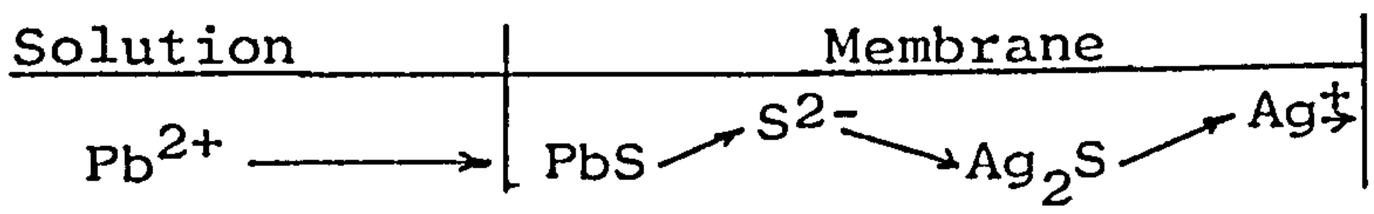
The response to lead is also much reduced from the Nernstian value, but is of equivalent magnitude with opposite sign to the response towards sulphate.

The membrane was used as an endpoint sensor in the titration of sulphate with lead or silver and in both cases the endpoint was indistinct. In the case of the lead titrant, the potential change was very small which, coupled with the noise in membrane potential, gave a large error. With a silver titrant the membrane potential changed by several hundred millivolts as soon as the first drop of silver was added and no inflection was observed at the expected endpoint.

15.3. Conclusion

The sulphate electrode, prepared as described by Rechnitz, has been found to give a poor response. It has a limited life time which is possibly caused by recrys talization within the membrane, and suffers from poor reproducibility. When used as an endpoint sensor, the membrane gives rise to indistinct or non-existent equivalence points and large titration errors.

It has been proposed that the lead selective electrode, based on silver and lead sulphides, responds through a reaction chain as follows (26):



15.3.-1

A possible explanation for the response of the Rechnitz membrane to the sulphate anion would be via the complex

reaction chain.

Solution	Membrane	
$SO_4^{2-} \longrightarrow$	SO_4 Pb $\nearrow Pb^{2+} \searrow$ PbS $\nearrow S^{2-} \searrow Ag_2S \nearrow Ag^+$	15.3.-2

If this is the case the crystal structure of the membrane must be of critical importance to its operation. Rechnitz (38) has shown that the presence of small amounts of sulphate in a lead electrode will impair the response or stop it functioning and has devised a complex washing procedure to eliminate it. If therefore the presence of sulphate can stop the reaction chain (15.3.1), it is quite likely that the reaction chain (15.3.-2) would also be interrupted unless a complex crystal structure made it possible. A time dependent breakdown of this crystal could explain the limited life time of the membrane.

From the observations made during the study of the sulphate membrane it is suggested that it is not at present useful as an analytical tool, and cannot be utilized as a sensor for automated analysis.

CHAPTER 16

The Development Of A Solid-State Reference Electrode

16.1. The Preparation of a Reference Electrode Liquid Junction Membrane

16.1.1. Preparation of the Membranes

16.1.2. Evaluation of the Membranes

16.1.3. Results and Discussion

16.2. The Solid-State Reference Electrode

16.2.1. Preparation of the Electrodes

16.2.2. Evaluation of the Electrodes

16.2.3. Results and Discussion

16.3. Conclusions

Ion selective electrodes can easily be made in solid state form, with no internal filling solution. These electrodes are very rugged and can be used in any position. Most available reference electrodes, however, consist of an electrode of the second kind immersed in an electrolyte contained by a liquid junction. Various methods of liquid junction formation have been used including agar gel connections, wicking, asbestos fibres, porous ceramic rods and many others. The junction behaviour must be substantially independent of the character of the test solution, and a method of evaluation of electrode liquid junctions using the Henderson equation has been devised by Light (123). Although the structures referred to above usually satisfy these conditions they are frequently slow and costly to make and suffer from high flow rates of reference electrolyte. The production of a solid state reference electrode would be very useful.

Apart from the obvious advantages of robustness and lack of solutions to be replaced, a solid state membrane could be machined to any desired shape,including flow through designs. Beckman have produced liquid junction structures, from various polymeric resins and inert materials that have proved to be reproducible and easy to manufacture (124,125). These junctions are designed to function by ionic diffusion rather than solution flow and are therefore less liable to give sample contamination. Liquidless junctions that can easily be manufactured and developed to provide an all solid state reference electrode could eventually be coupled with solid state indicating electrodes to form cheap disposable analytical devices. These disposable electrode systems could be utilized in body fluids and with radioactive substances,where contamination must be avoided. In this study liquid junctions were fabricated from both P.V.C. and P.T.F.E. in an attempt to produce an all solid state reference electrode.

16.1. The Preparation of a Reference Electrode Liquid Junction Membrane

16.1.1. Preparation of the Membranes

Five membranes were fabricated, using two basic materials, poly(vinyl-chloride)(P.V.C.) and polytetra-fluorethylene (P.T.F.E.).

I. A P.V.C. membrane was cast as for the immobilized exchanger electrodes, with the absence of any ion exchanger; 0.1g P.V.C. in a 1" I.D. glass ring.

II. 0.1g of P.V.C. was dissolved in 3 mls of T.H.F. that had previously been saturated with potassium chloride at room temperature. The membrane was cast in a 1" I.D. glass ring.

III. This membrane was prepared according to the Beckman Patent (125). Fine powdered P.T.F.E. (5μ diameter) and 10% by weight of KCl were ground together, made into a pellet 1.5 mm thick by a load of 15 tons in a 13 mm diameter infra red pellet press for one hour, and then sintered at 360^{o}C for one hour.

IV. A P.T.F.E./alumina (100-200 mesh) mixture, 1:1 by weight was prepared and treated as for membrane III.

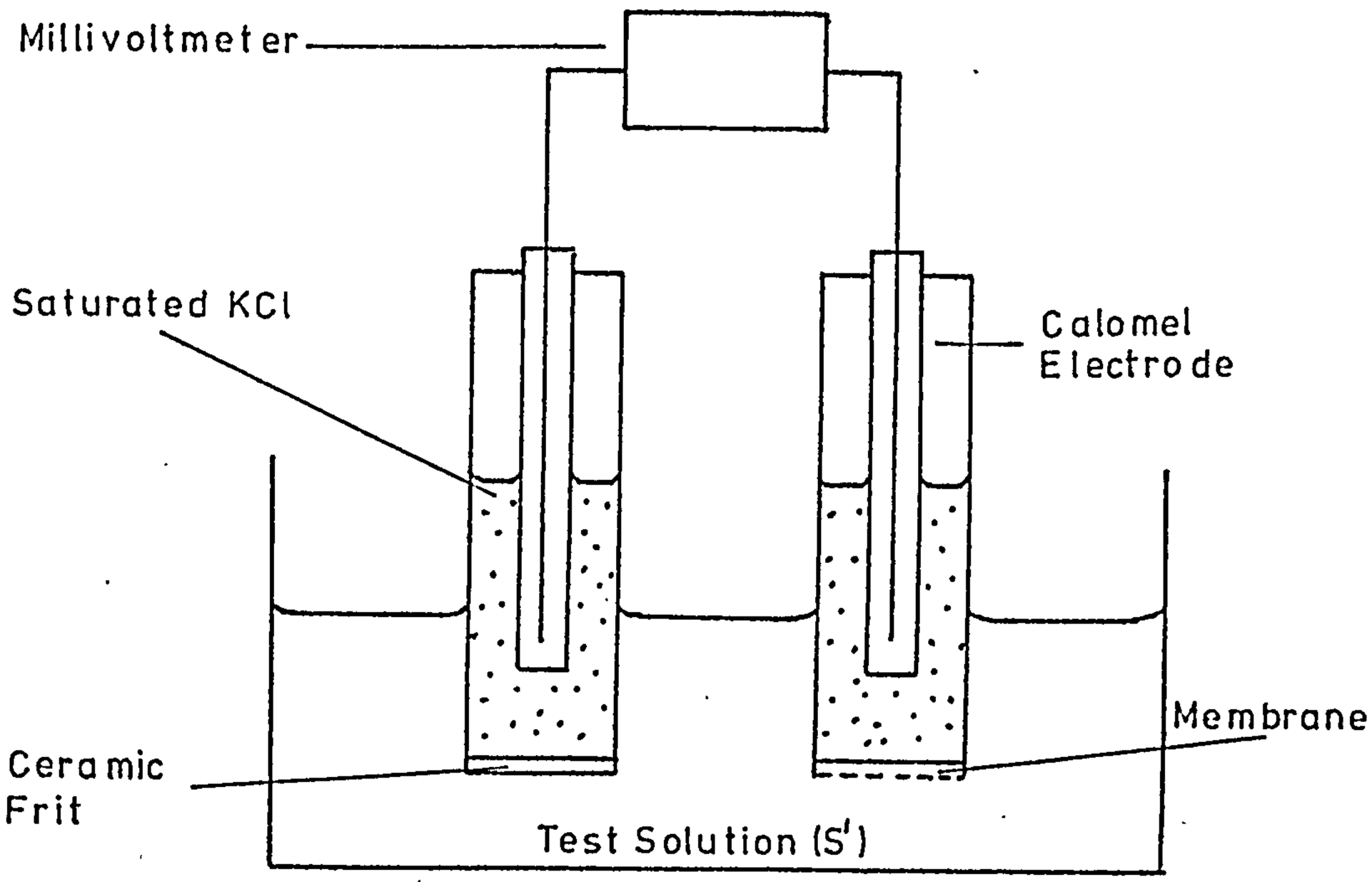
V. P.T.F.E. (5μ diameter) alumina (100-200 mesh) and KCl were mixed in the ratio 9:9:2 by weight. Sufficient mixture was treated as for membrane IV to produce a membrane of the same dimensions.

All membranes were mounted into P.V.C. tubes and soaked in distilled water overnight.

6.1.2. Evaluation of the Membranes

The P.V.C. tube and mounted membrane were arranged in the experimental design shown in Figure 16.1.-I. The calomel electrodes were connected to a millivoltmeter, with the electrode inside the ceramic frit acting as the 'reference'. The concentration of the test solution (S') was varied and the potential difference generated between the electrodes was recorded. In all cases the liquid junction potential of the ceramic frit, with a salt bridge of saturated KCl, was assumed to be constant, thus the potentials given in the results are the liquid junction

Figure 16.1.-I



Experimental Arrangement for Membrane Evaluation

potentials of the membrane 'relative to a ceramic junction', and are theoretically zero.

In order to check the response of the 'membrane junction' reference electrodes in the absence of any other liquid junctions, they were used as reference membranes in conjunction with glass electrodes to measure the pH of several standard buffers.

16.1.3. Results and Discussion

Membranes I and III proved to have such a high resistance that the voltmeter registered open circuit for all measurements. Membrane III was reduced to 0.5 mm thick, but without any improvement. The results for the other membranes are tabulated below (Table 16.1-II).

Table 16.1-II Reference Junction Potentials (mV)

Membrane Solution		II	IV	V
KCl	Sat (4.1m)	+0.3	+0.3	0.0
	2.5 M	+0.0	+0.5	0.0
	1.25	-0.5	+1.9	+0.3
	0.1	-0.5	+27.0	+0.4
	0.01	-16.9	+41.7	+4.0
$NaNO_3$	2 M	-0.7	+0.3	-0.1
	1	-1.6	+1.3	0.0
	0.1	-6.2	+17.9	+2.2
	0.01	-15.8	+47.0	+3.0
pH	0.75	0.78		0.74
Buffer*	3.76	3.99		3.79
	7.00	7.00		7.00
	9.12	9.01		9.06

pH value quoted obtained from calomel/frit electrode as reference.

*pH 7 buffer used as standard to set all electrodes on scale.

If the ceramic frit of the calomel reference electrode half-cell is assumed to be a good liquid junction, then a membrane behaving in a similar fashion would have the same potential difference across it, and in the experimental arrangement shown in Figure 16.1-I a zero potential would be indicated between the two calomel electrodes. With the membranes made in this study, it can be seen from the results that those approaching this situation are membranes II and V.

The effects of potassium chloride in the membranes are significant, but the salt itself is not a guarantee of membrane operation. In the P.V.C. membrane (II) the matrix is made more hydrophilic by the presence of even a little salt and the membrane becomes operable. Membrane III however contained 10% of potassium chloride, and did not operate. This concentration of salt is not sufficient to allow particle contact and as the bulk of the membrane was composed of a hydrophobic, non-ionic polymer,neither ionic nor solution diffusion could take place. When 50% alumina is added to the P.T.F.E. the membrane becomes permeable to ionic species, due to the ease with which it can be hydrated. The addition of the ionic salt (KCl) to the mixture also aids diffusion through the membrane and the junction potential is considerably decreased. A greater concentration of salt in the membrane might decrease the junction potential further, but there is an increased risk of membrane dissolution and formation of pores through the membrane bulk.

The pH response of the membranes containing alumina can be related to the materials' activity. The alumina available had a Brockman activity of 1-2, and a more neutral, hydrophilic material might decrease the response to pH.

Both membranes II and V could be used as reference junctions but,of the two,membrane V is the most promising as constant voltage electrode, whereas membrane II, applied as a coating, could provide a good micro reference electrode.

16.2. The Solid State Reference Electrode

The membranes previously tested were adapted and examined as solid state reference electrodes.

16.2.1. Preparation of the Electrodes.

VI. Sufficient mixture of alumina, teflon and potassium chloride (9:9:1 as for V) was placed in a pellet press (Figure 5.2.-I) to provide a pressed thickness of one millimeter. More mixture, increased to 30% of KCl by weight, was placed on top, a silver/silver chloride wire inserted and the whole was compressed at 15 tons for one hour. The pellet obtained was then sintered at 360°C for one hour, mounted into a P.V.C. tube with epoxy adhesive and soaked in distilled water overnight.

VII. The end of a silver wire 0.5 mm in diameter was melted in a flame to form a small drop. This was cleaned and coated with silver chloride electrolytically and subsequently dipped in a P.V.C./T.H.F.(KCl) mixture (as for membrane II). The small drop on the end of the wire ensured that there were no sharp points from which the P.V.C. coating could easily break away. The electrode was left 48 hours to dry and then soaked in saturated potassium chloride solution overnight. Electrodes were also prepared from wires dipped in P.V.C./T.H.F. without KCl, and an uncoated silver/silver chloride wire.

16.2.2. Evaluation of the Electrodes

After washing with distilled water the electrodes were tested for stability by recording their potentials with respect to a calomel reference electrode over a period

of twelve hours. The electrodes were then introduced to solutions of different ionic strength and various pH buffers to evaluate them as reference electrodes.

As an application to analysis the electrodes were used as references in the titration of alkali with acid, to a glass electrode potentiometric endpoint.

16.2.3. Results and Discussion

The potential drifts of the electrodes were evaluated in 0.1 M sodium nitrate; electrode VI, -0.1 mV/hour, electrode VII, + 1.8 mV/hour.

The effects of solution strength and pH are tabulated below (Table 16.2.-I).

Table 16.2-I

Solution	Electrode VI	VII Ag/AgCl/PVC,KCl	Ag/AgCl/PVC	Ag/AgCl
KCl Sat	-71.9	+203.3	-333	+72.8
2.5	-70.9	+197.2	-368	+116.4
1.25	-70.1	+186.8	-379	+79.0
0.1	-68.8	+168.3	-420	+19.3
0.01	-66.9	+168.5	-500	-28.0
KNO_3 2M	-67.5	+187.2		
1M	-67.4	+186.7		
0.1	-66.6	+186.2		
0.01	-64.1	+184.0		
pH* 1.06	1.30	1.17		
4.16	4.18	4.21		
7.00	7.00	7.00		
8.88	8.90	8.75		

* established with a glass electrode and calomel electrode with a sat KCl bridge and a ceramic frit. pH 7.00 adjusted as standard.

Reference Membrane Potentials (mV)

From these results it can be seen that the P.V.C./KCl coated wire electrode acts as a good reference in a solution of nitrate ion but responds to some extent in solutions of chloride ion, although with a greatly reduced response in comparison to an uncoated silver, silver chloride wire. The response in solutions of different pH is poor. Electrode VI behaves in a similar fashion to the junction membrane V but has poorer pH response.

The titration curves of acid vs. base to a potentiometric endpoint using the solid state reference electrodes are shown in Figure 16.2.-II. These curves indicate that of the solid electrodes prepared, electrode VI gives the response that most resembles the calomel electrode with a ceramic frit. The inflection point of the curve corresponding to the P.V.C. coated silver wire showed a change in position, indicating a response to the added chloride ions. There is no decrease in the potential jump with this electrode, however, that is associated with the uncoated silver/silver chloride wire.

16.3. Conclusions

Although it has been shown that all-solid-state reference electrodes are feasible, inexpensive and easy to manufacture, much more work is necessary to produce a reliable interference free electrode. The pH dependence of the pressed pellet electrode could be improved by using an intrinsically neutral inert material, or neutral mixture of acid and basic materials (e.g. Silica + alumina) as the hydrophilic additive to the P.T.F.E. matrix.

E mV vs.
Glass pH Electrode

100

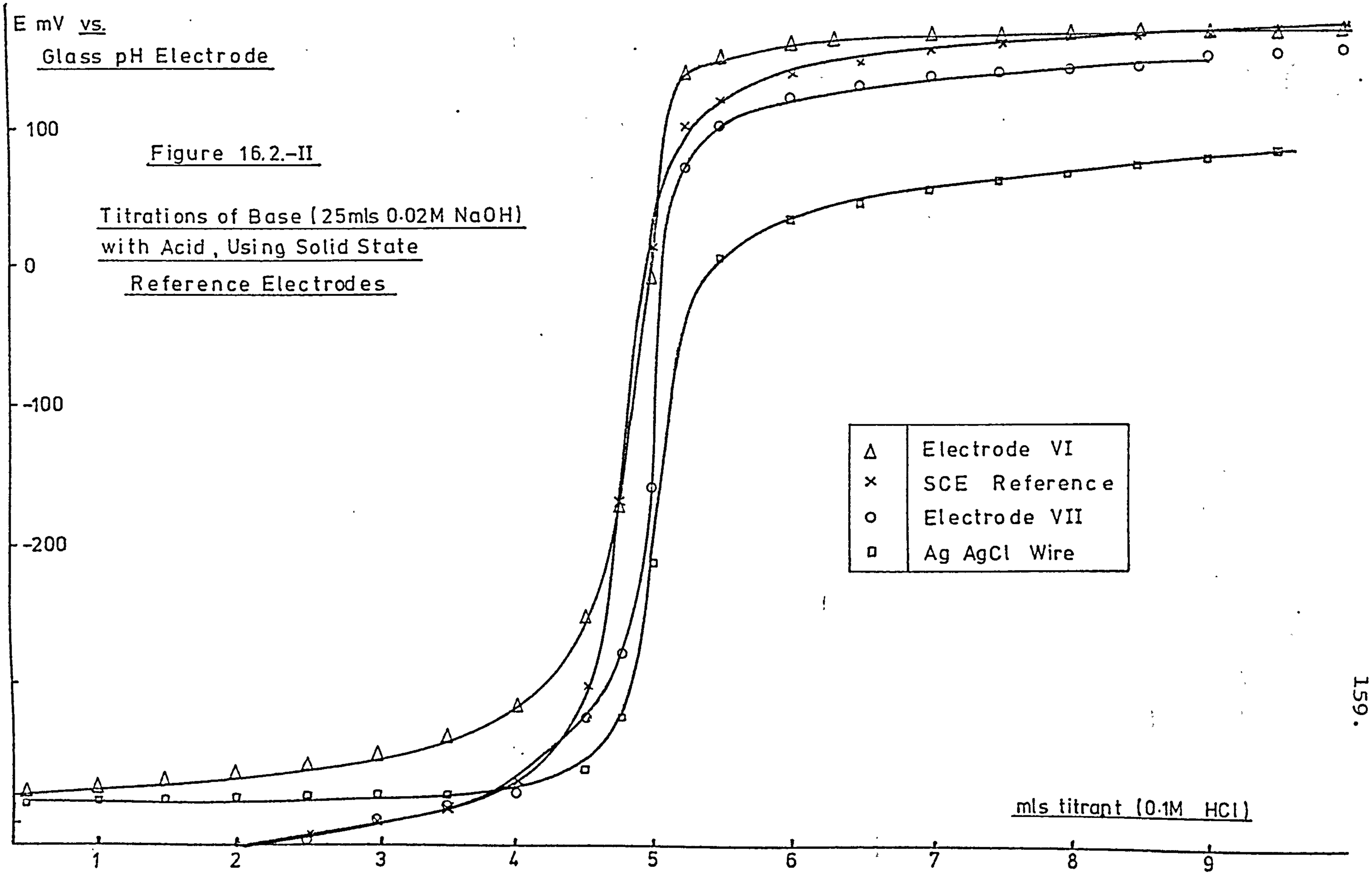
0

-100

-200

Figure 16.2.-II

Titrations of Base (25mls 0·02M NaOH) with Acid, Using Solid State Reference Electrodes



The major problem with the coated wire electrode seems to be the absence of a constant-chloride environment at the silver surface. A T.H.F. solution of poly(vinyl-chloride)is not able to contain sufficient potassium chloride to provide a constant level for an extended period and so other polymers must be investigated for this purpose. Poly(vinyl-alcohol) can be used as a matrix to immobilize a salt in an aqueous solution but subsequent coating with a poly(vinyl-chloride) layer is difficult and is liable to rupture by osmotic pressure. Until the problems of polymer coatings can be solved, the pressed pellet solid state reference electrodes can be used, and offer considerable promise for the future.

SECTION III

Automation of Ion Selective Electrode Potentiometry

CHAPTER 17

Automated Techniques, Using Ion Selective Electrodes As Sensors

17.1. Automation of the Analytical Technique

17.1.1. Sampling

17.1.2. Sample Processing

17.1.3. Sensing

17.2. The Choice of Analytical Technique Using Ion Selective Electrodes as Sensors

17.2.1. Standard Additions - Gran's Plot Modification for Continuous Flow Streams

17.2.2. Automated Titration to a Potentiometric Endpoint

17.2.2.1. The Gradient Titration

17.2.2.2. The Single Cup Gradient Titration

17.2.2.3. The Single Cup Gradient Titration with Coulometric Generation of Titrant

The International Union of Pure and Applied Chemistry (IUPAC) refers to automation as "the use of combinations of mechanical and instrumental devices to replace, refine, extend or supplement human effort and facilities in the performance of a given process, in which at least one major operation is controlled, without human intervention, by a feed-back system". Although many so called 'automated techniques of analysis' do not come within this strict definition and should really be termed 'mechanised techniques', it is generally agreed that an 'automatic method' implies the elimination, partially or fully, of human intervention in a chemical method (126).

Recent development of automated methods of analysis (126-132) has followed progress in three main fields; the increase in multi-element study in clinical laboratories, cost consciousness of industry necessitating rigid control of the manufacturing processes and pressures caused by the increasing public awareness of environmental pollution.

Within the past fifteen years larger hospital populations, an increase in the use of preventive medicine and increased reliance on chemical analysis for diagnosis have been responsible for the increase in workload on the biochemical and hospital laboratories. To cope with the heavy workload and necessarily rapid turnover in analysis, automated instrumentation must be used. The continuing increase in automation in clinical biochemistry is indicated by numerous reports in the literature concerning new and improved methodologies and their clinical applications (127-135). The increased capabilities of the clinical laboratories as a result of automation has caused a demand for analysis of a whole range of body fluid constituents to provide profiles of individuals under study (136).

The continuing increases in world prices of raw materials has meant that industrial processes must be controlled within very fine limits to avoid any waste. The high volume output of modern industrial plants, and the strict requirements on the quality of the end products require instruments to provide dynamic rather than historical analysis of the process stream, so that remedial action can be effected. This constant surveillance at each process step can only be readily achieved by continuous monitoring analysis,

or, less satisfactorily by rapid repetitive analysis of the individual process steps. Agricultural analysis, although not yet at the pressure of its industrial counterparts, is increasing rapidly and the analysis of soils, fertilizers, feeds, plant tissue and farm effluents is a field which is benefiting from the use of automated procedures.

Increases in population density, traffic and industrialization have made pollution control a complex problem, (137). An initial step in any pollution control programme must be the establishment of realistic clean air and clean water standards, requiring a large volume of background data. The acquisition of such data and the monitoring of possible sources of pollution, such as industrial wastes, is a simple application of an automated analytical system, of which there are at present many commercial models on the market.

17.1. Automation of the Analytical Technique

When considering the automation of an existing manual analytical technique the main factors to be determined are a saving in time, (in relation to the number of determinations needed), the reduction in cost of individual analyses, a possible increase in accuracy and precision and the initial cost of the automating equipment. However, when the number of samples is very large, it becomes impossible to provide a quick turn around without the facility of a reliable automated technique.

The sequence of an automated analytical technique can be divided into three main sections; sampling, processing and measurement.

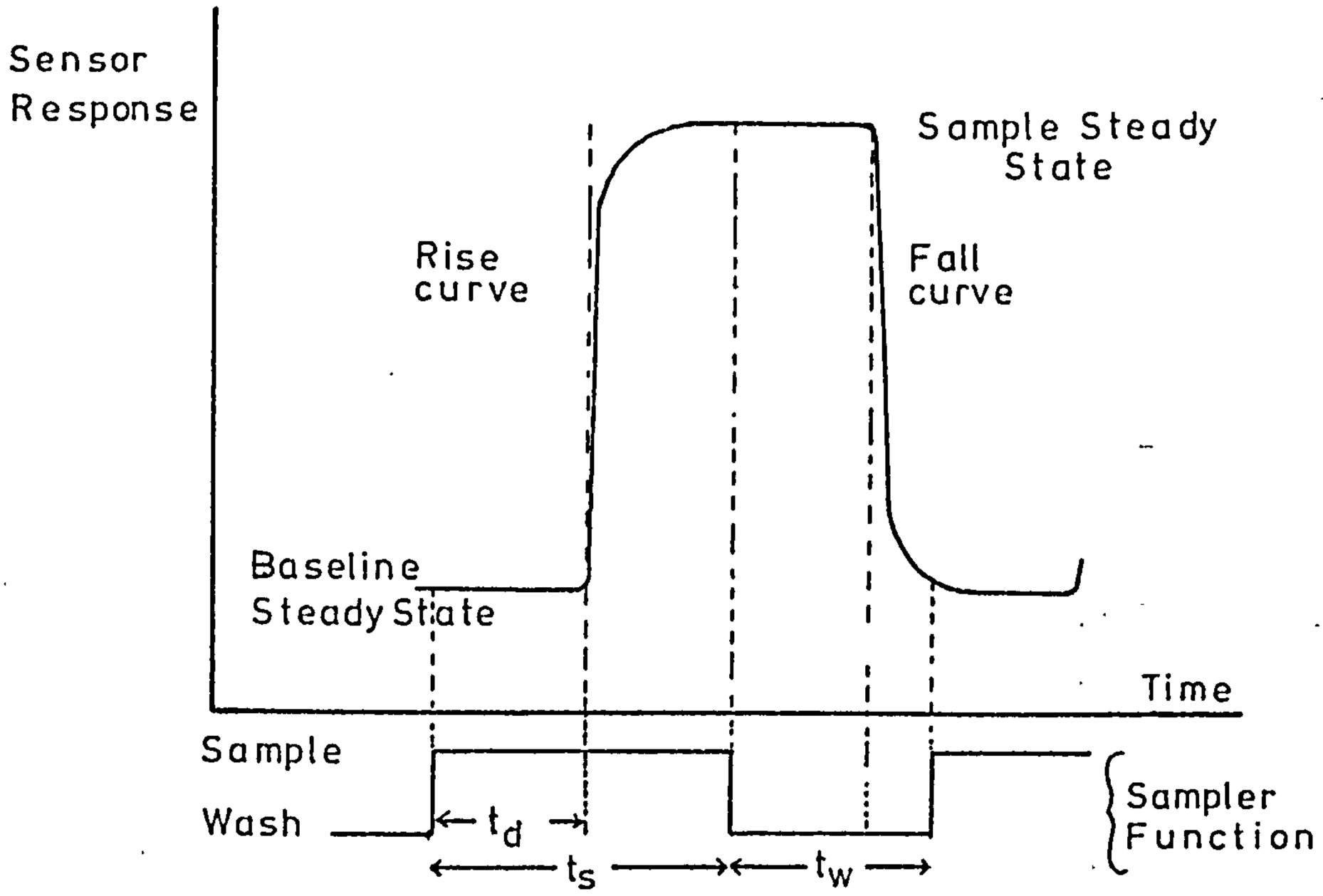
17.1.1. Sampling

Although some selective electrodes are capable of sensing the concentration of a species in the gaseous phase, the majority respond only to ions in solution. Solid samples must therefore be dissolved before they can be processed and this can be the biggest hurdle to overcome in an automated procedure. Metal samples, however, have been prepared using anodic dissolution (138,139) and Technicon produce a 'Solid-Prep' unit which is a high-speed homogeniser for dissolving samples such as fertilisers and pharmaceutical preparations. Sampling from industrial process streams, where the analyte is already in solution, can be done simply by bleeding a proportion of the sample through a flow controller to the analytical unit. Analysis can then be done either continuously or whenever the controller is activated. Batch samples of liquids are converted into a flow-stream by pumping from a series of sample cups, interspersed with wash to prevent cross-contamination and to aid sample identification. The latter system is used in this work in conjunction with Technicon Auto Analyzer pumps and samplers. The concentration changes during the sample and wash cycle have been thoroughly investigated by several workers (140,141,142) and the treatment and terminology given here are similar to that of Thiers and coworkers (141).

Figure 17.1.-I shows the concentration profile at the sensor as a function of time, normally obtained with continuous flow analysers. When a blank (or wash) solution is being sampled, the concentration corresponds to a 'baseline steady state'. The inlet probe is then placed in a sample at the beginning of the 'sampling time' (t_s). The sensor does not observe this change in concentration until the sample has travelled along the pump tube from the sampler to the sensor in a time t_d. This is known as the 'dead time', or 'delay time'. At the end of t_d the sensor will respond to the sudden increase in concentration along a line called the 'rise curve', and if the sampling time is sufficiently prolonged, a stable reading will be obtained at the 'sample steady state'. At the end of t_s, the sampler probe is transferred to the wash solution and forms the beginning of the 'wash time', (t_w). After a further period, t_d, the effect of this change will be observed at the sensor and the response will decrease along the 'fall curve' until, if t_w is long enough, the baseline steady state is reached. The sum of t_s and t_w then determines the sampling rate. If the times are kept too short the response may not reach the sample steady state, resulting in low values, and the baseline steady state may not be reached giving rise to 'sample carry over' or 'sample interaction'. These problems are usually avoided by keeping t_s and t_w sufficiently long to ensure that the steady state values are reached but some work has been done on a dynamic model for continuous segmented flow

analysis (142,143) which provides some insight into the development of more accurate techniques of non-steady state analysis.

Figure 17.1.-I



— Sensor Response to Concentration Profile as a Function of Time

17.1.2. Sample Processing

Sample processing consists of preparing the analyte for sensing and measurement. This may include buffering, mixing with reagents, heating, dialyzing etc., most of which can easily be carried out by incorporation of the appropriate 'Technicon' or similar module in the analysis manifold. Concentration gradients can also be generated and used to perform automated potentiometric titrations. This method of analysis, known as the 'Gradient titration' will be fully described later.

17.1.3. Sensing

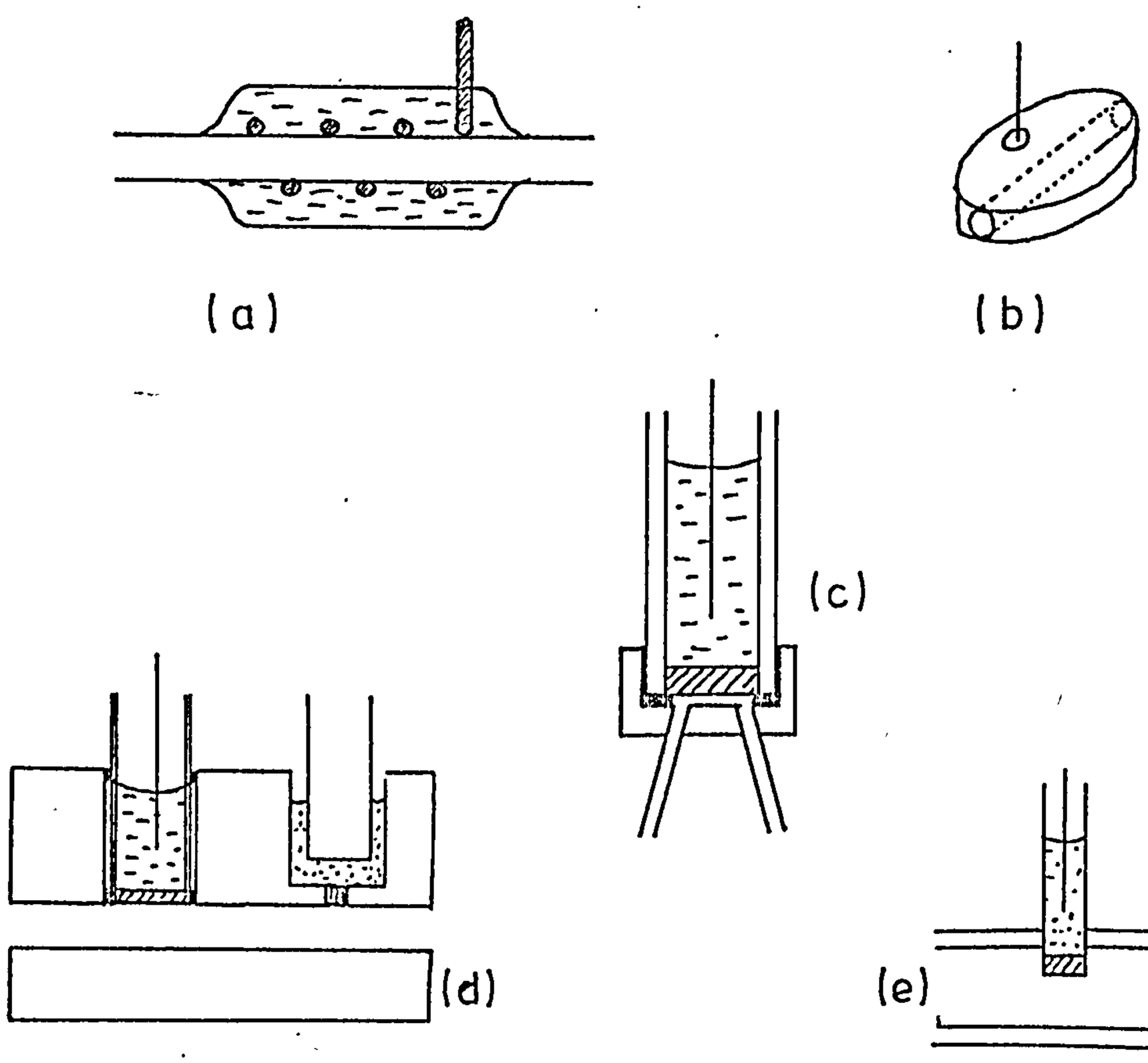
Sensing or measurement, consists of detecting a certain property in the processed sample which can be correl ated with the composition of the sample. A comprehensive review of the applications of various sensors to continuous analysis has been presented by Blaedel et al.(127). The types of continuous sensors reported cover almost the entire field of physico-chemical analytical techniques, including visible spectrophotometry, flame photometry, infrared and ultra-violet spectrophotometry, emission spectroscopy, atomic absorption spectroscopy, atomic fluorescence spectroscopy, mass spectrometry, amperometry and other electrochemical techniques including ion selective electrodes.

There have been several reports of ion-selective electrode applications in continuous analysis, fluoride, (144), cyanide (75) and sulphide determinations provide examples of the general approach to the problem. One of the main difficulties in the design of a continuous analysis system is the choice of measuring electrode and reference electrode. This problem can be formidable when one considers that the original main classification of the I.S.E. into glass, solid state and liquid membrane electrodes must take into account many sub-classes. The copper electrode for example has no less than seven different configurations; including glass, solid state mixed crystal, heterogeneous immobilized and liquid membrane electrodes. The final choice must depend on the application involved, but reliability, expertise of the operator and economic factors are all relevant.

The electrode design in continuous analysis must of course allow for solution flow (Figure 17.1.-II). Most adaptations of ion-selective electrodes are carried out by fitting some form of flow-cap over the membrane but the capillary glass electrode (145,146) has found wide application in industrial and clinical areas of analysis, and a similar design has been employed with solid state membranes, where the membrane is drilled to allow solution flow.

Figure 17.1.-II

Design of Flow Through Electrodes



(a) Capillary Glass Electrode
(b) Drilled Solid State Electrode
(c) Flow Cap
(d) Flow Cell
(e) Direct Probe

Many problems in ion-selective potentiometry can be traced to a faulty reference electrode. In this respect continuous monitoring offers an important advantage since the reference electrode, once it is functioning, remains in a constant environment. There have been various approaches to the problem of reference junction design. Capillary junctions with a continuous slow flow of reference electrolyte have been found useful but the recently described immobilized P.V.C. and Teflon (125) reference electrodes have been tried and offer considerable promise.

Almost all electrodes have dependence of electrode potential on flow rate. The calcium electrode for example, shows an initial sharp increase in potential and then a plateau region with increasing flow rate. The initial increase is due to the electrode potential involvement with the membrane solution double layer. The flow rates used in normal analysis, however, are in the plateau region and the potential is independent of the minor fluctuations in flow encountered.

The speed with which samples can be processed is obviously important in an automatic system and is frequently controlled by the response time of the monitoring system. While the real response times of glass and solid state membranes in pure solutions are of the order of milliseconds and should not significantly influence most measurements, those of liquid membranes are of the order of seconds. In real situations when the influence of interfering ions and the contribution from hydrodynamic mixing are taken into account, response times of 15 secs.

to several minutes may be observed for the liquid membrane electrode. This time can be cut down by using the potential at a known proportion of the potential change, i.e. t_{95} (67). This proportion of the equilibrium should be well defined after a period of about 20 seconds except in the most adverse conditions.

17.2. The Choice of Analytical Technique Using Ion Selective Electrodes as Sensors

The choice of analytical technique is similar to that encountered in manual analysis vis.

a) Direct potentiometry.

b) Direct potentiometry with precalibration.

c) Direct potentiometry with interposed standards.

d) Standard additions - Gran's plot modification for continuous flow streams.

e) Automated titration to a potentiometric endpoint.

In automated analysis, direct potentiometry, direct potentiometry with precalibration and direct potentiometry with interposed standards have the same merits and demerits associated with them as for manual analysis.

17.2.1. Standard Additions - Gran's Plot Modification For Continuous Flow Streams

The method of analysis using multiple additions to a known volume of sample solution has already been described (8.1.5). The technique can be modified to automated analysis using the experimental design shown in Figure 17.2.-I. A continuous sample stream is maintained at a flow rate of 2F mls/min. It is mixed with a continuous

buffer flow at Fml/min and a series of standards sequentially presented at F ml/min separated by a water wash. When the junction potentials and activity coefficients are assumed constant, the electrode potential can be represented by equation 17.2.1.-I

where volume is replaced by its flow rate equivalent:

$$E_i = \text{Constant} + S \log \frac{2FC_o + F\,C_i}{4F} \qquad 17.2.1.\text{-I}$$

$$= \text{Constant} + S \log \frac{2C_o + C_i}{4} \qquad 17.2.1.\text{-2}$$

Where E_i is the potential of the system where the concentration of the standard addition stream is C_i and C_o is the concentration of the sample test solution.

The equation can be rearranged to give:

$$10^{E_i/S} = 10^{\text{Const}} \times \frac{2Co + C_i}{4} \qquad 17.2.1.\text{-3}$$

A plot of $10^{E_i/S}$ vs. C_i will give a straight line which intercepts the abcissa at C_A, such that

$$CA = -\,2C_o$$

A computer program 'AUTOGR' has been written in Fortran IV (147) to evaluate the unknown concentration from the input data, cell potential and concentration of the addition standards.

17.2.2. Automated Titration to a Potentiometric Endpoint

Currently available automatic potentiometric titrators can be divided into two main classes; i) the 'batch' titrator, in which a measured titrant volume is added to a sample solution with a recorded titration curve and ii) 'continuous' titrators, where a fixed flow rate of sample is mixed with

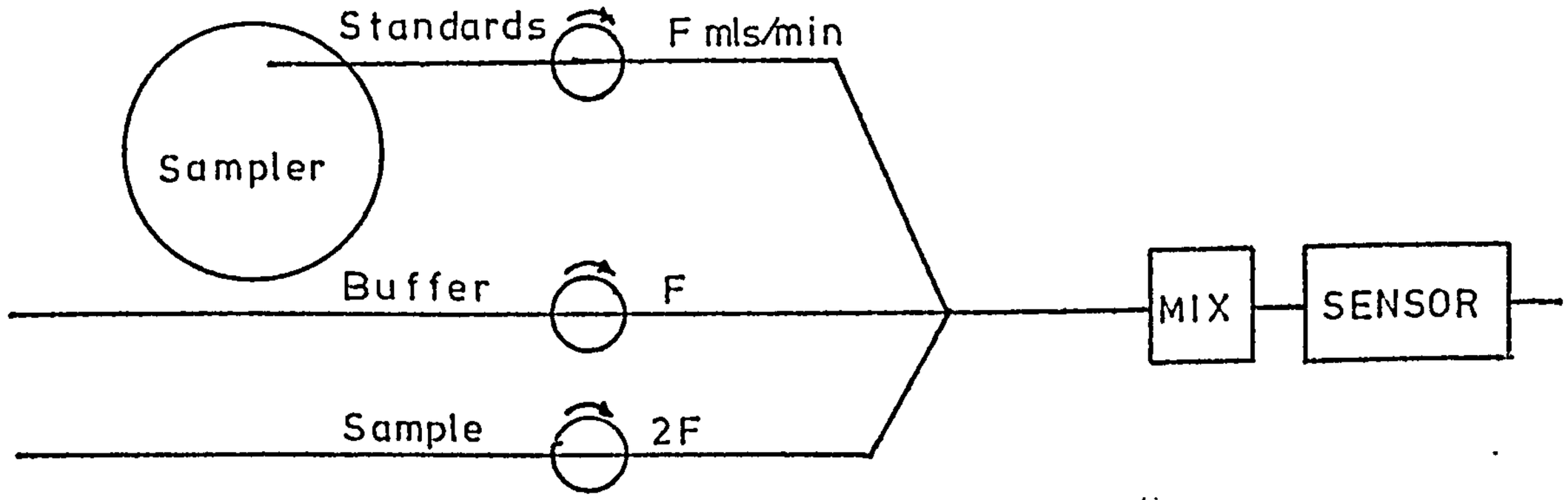
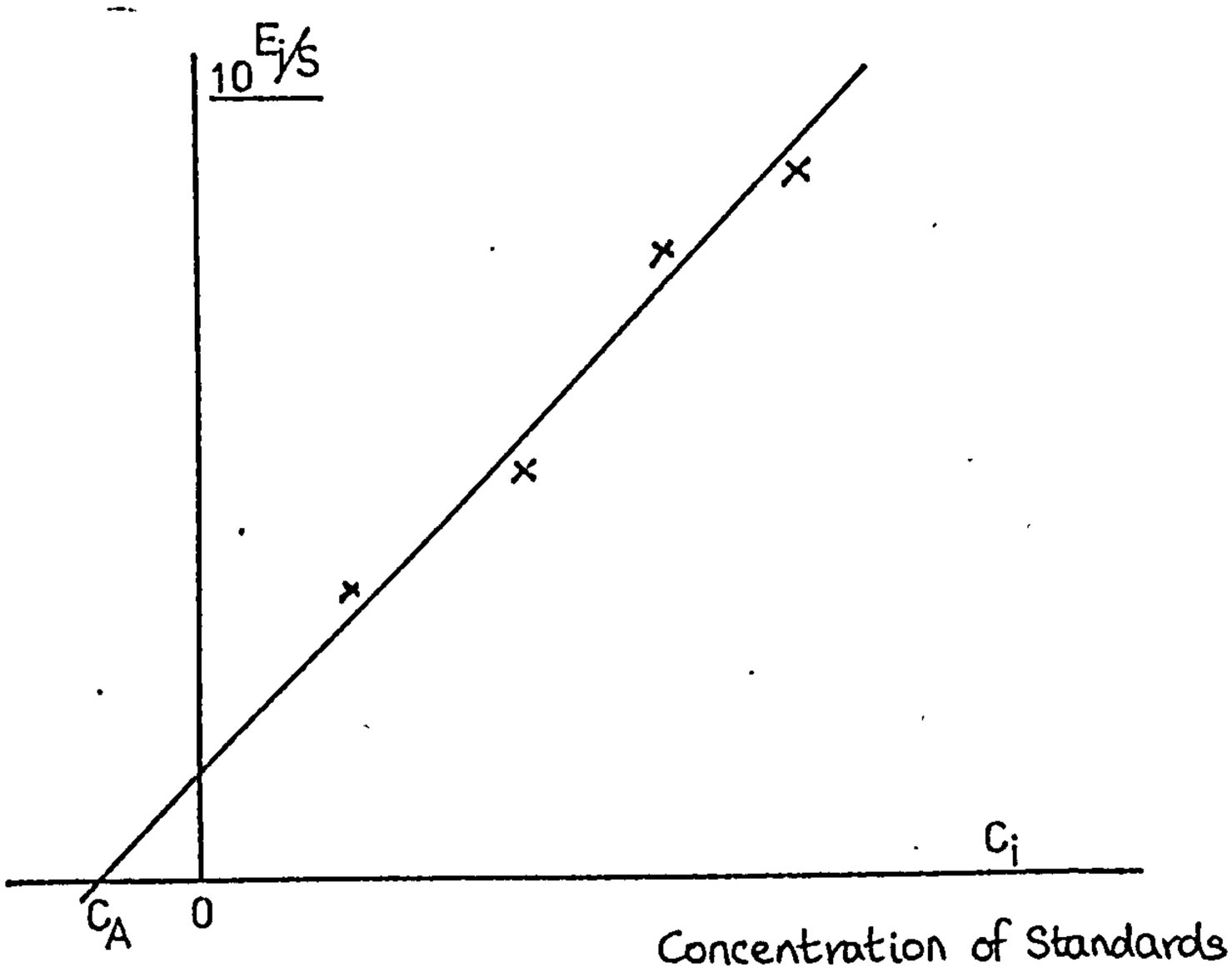


Figure 17.2.-I

Continuous Analysis Modification of the Gran's Plot Procedure



titrant, whose flow rate is varied until equilibrium conditions are satisfied, and the sample concentration is calculated from the titrant flow rate. Both of these titration systems have been produced with a digital readout facility (148,149) and are comprehensively reviewed by Foreman and Stockwell (126) and Buck (4,5,6).

17.2.2.-1 The Gradient Titration

An additional titration technique has been described (150), which combines the two previous techniques, allowing the titration of both continuous streams and batches of samples. It is known as the 'gradient titration'.

In classical titrimetric procedures the volume of titrant in a closed stationary system (titration vessel) is the variable which is controlled, monitored and subsequently used to calculate the original analyte concentration in the test solution. Another way of obtaining a controlled but variable amount of reagent (or titrant) is by means of a gradient. Concentration gradients have been used for a long time in liquid chromatography (151), and are easily produced with peristaltic pumps. If a flow of titrand (or sample) solution is mixed with a flow of titrant (or reagent) whose concentration is in the form of a gradient, one can visualize titration results similar to classical titration with a burette, but at constant volume. If there are two containers I and II containing solutions of different concentrations initially and fluid is being pumped from I to II and on from II, (Figure 17.2.-II) the concentration of B (C_B) at any instant is given by, (152),

$$C_B = C_A + \left[\frac{C_{BO} - C_A}{V_B}\right]\left[V_B + (F_A - F_B)\ t\right]^{\left(\frac{F_A}{F_B - F_A}\right)} \qquad 17.2.2.\text{-I}$$

Where C_A = concentration of A, C_{BO} = original concentration of B, V_B = original volume of II, F_A = flow rate from I to II, F_B = flow rate from II, t = time.

If F_B is madeequal to $2F_A$ the equation reduces to:-

$$C_B = C_A + \left[\frac{C_{BO} - C_A}{V_B}\right].\left[V_B - F_A t\right] \qquad 17.2.2.\text{-2}$$

The concentration of B is then a direct linear function of time. Figure 17.2.-II also shows the effect of using either A or B as the titrant, the concentration gradient either increasing or decreasing with time, until the stoichiometric equivalence point is reached at time T. The concentrations of any of the species may be monitored by ion selective electrodes to find the 'end point' at time T.

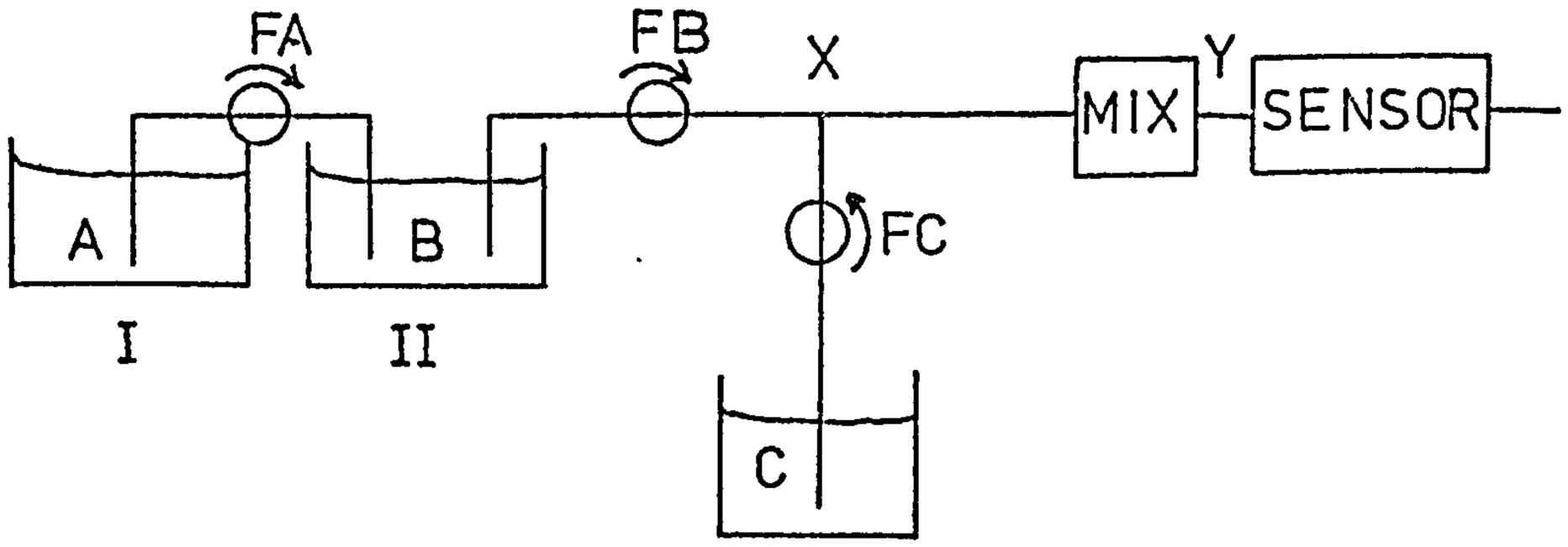
In addition to the gradient titration system being a precise analytical technique, it is also inexpensive. Apart from the gradient producing system (typically Technicon peristaltic pumps) the electronic equipment needed for the titrations consists only of a voltage following operational amplifier and a recorder. There is no need for an expensive digital volt meter. The system can also be applied to almost any titration, where an I.S.E. is available responsive to either the titrant, titrand or product solution.

17.2.2.2. <u>The Single Cup Gradient Titration</u>

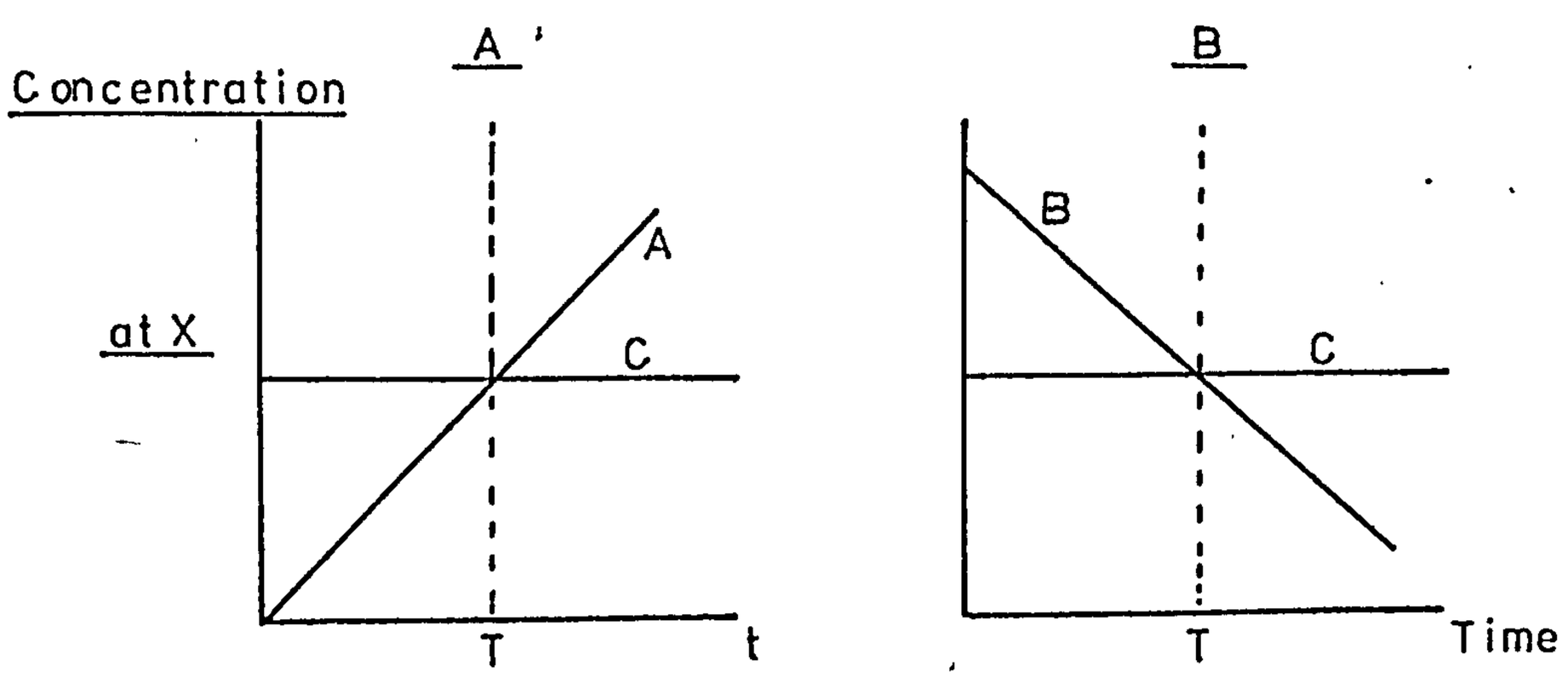
During this study, a simple method of automating a series of titrations has been developed which does not involve any modification of standard Auto Analyzer equipment.

Figure 17.2.-II

Gradient Titration



Reactant Species



In the simplest approach to gradient titration a constant flow of sample is mixed with a constant flow of reagent where the concentration of reagent is increased linearly in the form of a gradient. The gradient can be generated either by a 2 cup mixing technique or alternatively by coulometric generation. The variant described here was devised specifically for use with the Auto Analyzer system and involves the development of a reagent gradient within the sample container.

In the initial applications of gradient titration (Figure 17.2.-III), vessel I contains reagent, vessel II distilled water and the developing reagent gradient is pumped onward from II. If, however, vessels I and II contain different species, say A and B, then the concentration time profiles of A and B in vessel II will show a decreasing gradient of B and an increasing gradient of species A (Figure 17.2.-IV). In the case, for example, where I contains reagent and vessel II the sample solution, then the equivalence point will be reached during the titration at the time 'x' when the stoichiometric concentration ratio is obtained (Figure 17.2.-IVa). With only one reagent concentration (C_A) any concentration of sample (C_{BO}) may be analysed (Figure 17.2.-IVb). The relationships between solution flow rates, sample and reagent concentrations, sample volume and titration end-point are as follows:-

$$C_{Bo} = C_A.x\,/(D-x)$$

$$D = V_{Bo}/(F_B - F_A)$$

where x is the time elapsed between initiating the gradient and the endpoint, and D is the total titration time.

Figure 17.2.-III

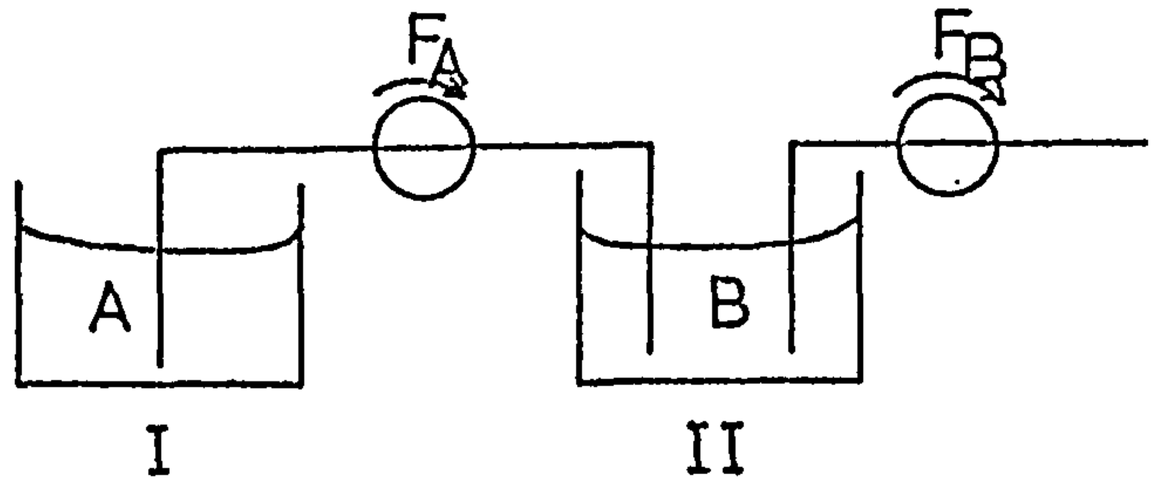
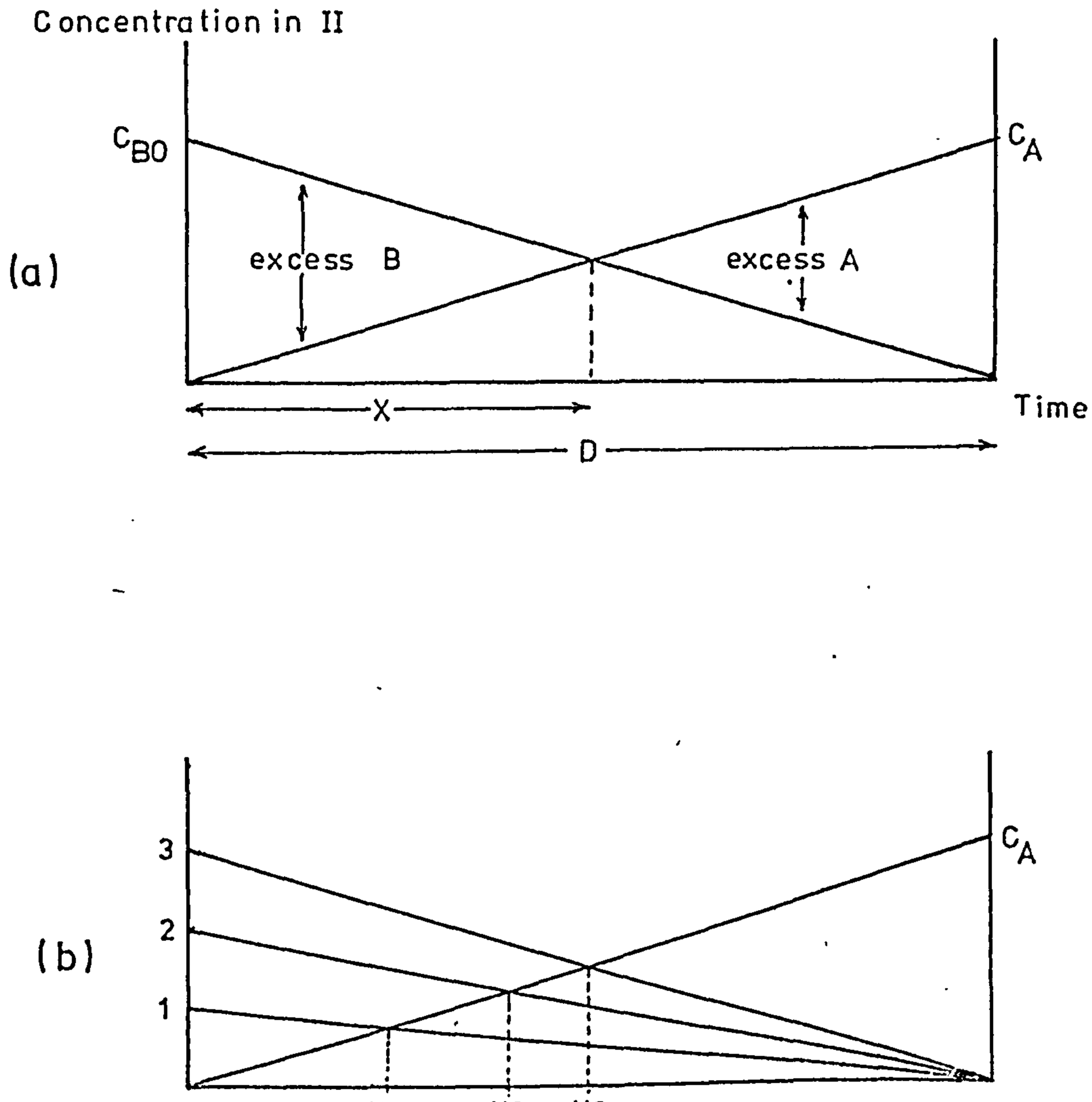


Figure 17.2.-IV

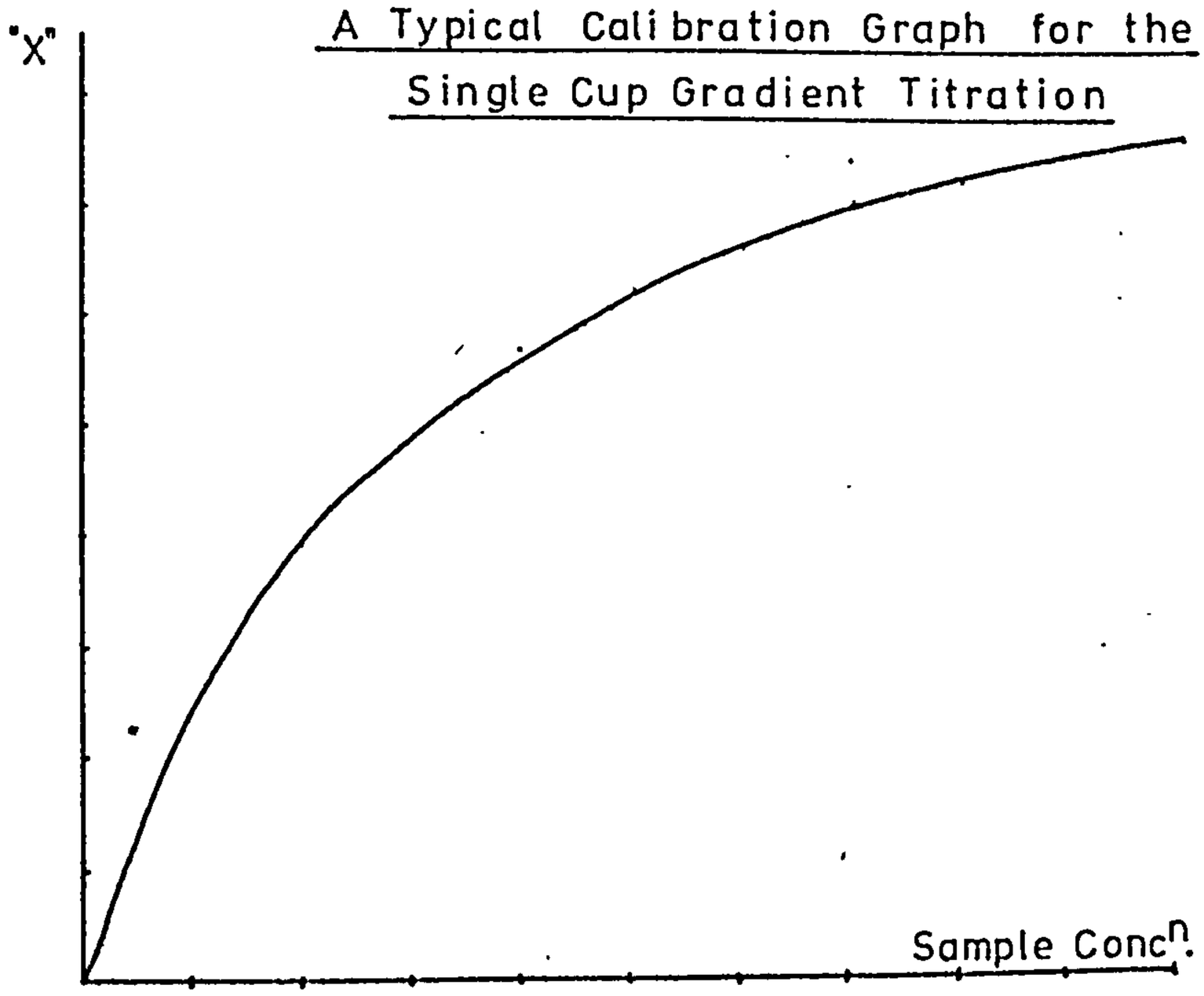


The Single Cup Gradient Titration Technique

F_B and F_A are the flow rates of solution out of II and from I to II respectively. As in the case of the two cup titration system, F_B must be equal to $2F_A$ to ensure a linear concentration gradient.

It can be seen from this treatment that a calibration graph of time(x) vs. concentration of sample (C_{Bo}) will not be linear; an example of a typical calibration graph is given in figure 17.2.-V.

Figure 17.2.-V



17.2.2.3. Gradient Titration With Coulometric Generation Of Titrant

Coulometric generation provides an elegant and very precise method of gradient production for those cases where the reagent forms part of a suitable electroactive couple. A schematic diagram of an experimental assembly for the coulometric titration is given in Figure 17.2.-VI. In the simplest case a single pump line is used with generation of reagent directly into the sample stream.

If the sample is electroactive at the generating electrode, however, then the reagent can be generated in a parallel line before mixing with the sample stream. The counter electrode can be located either in the sample stream, isolated by a porous junction, or in a line separated by dividing the stream into two, immediately before the measuring electrode.

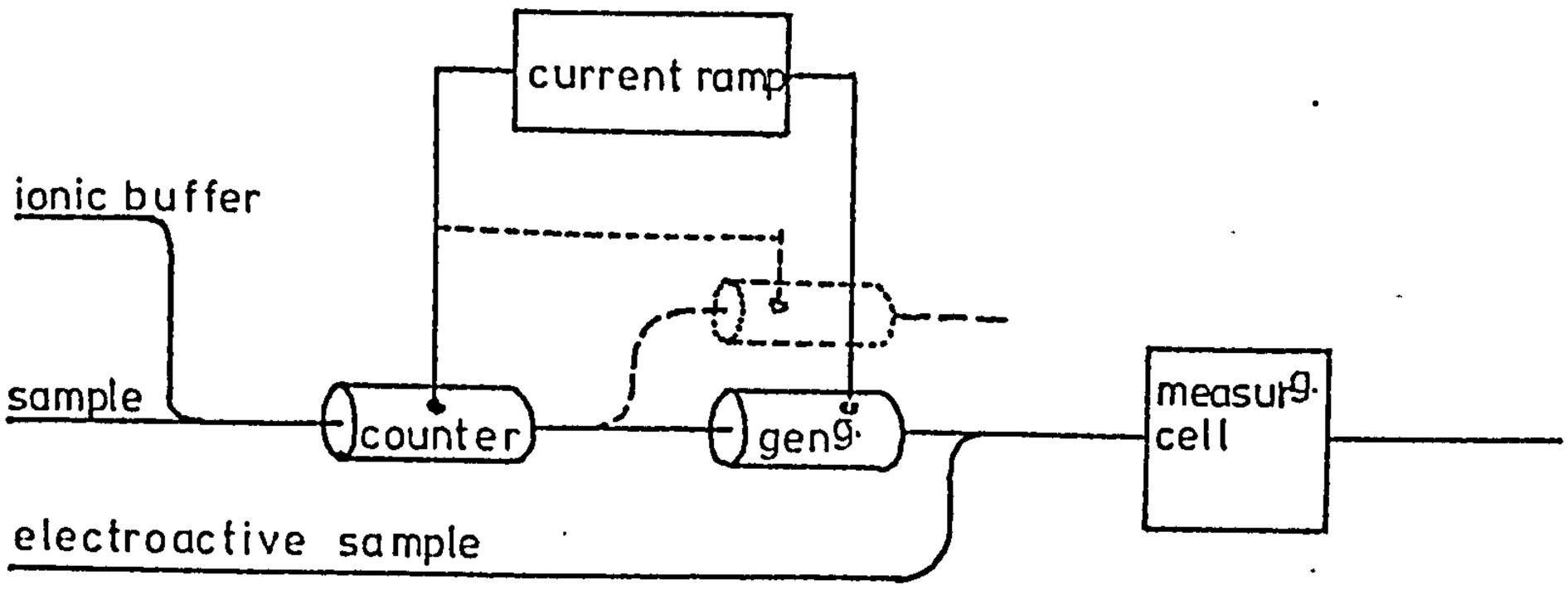


Figure 17. 2.–VI

Schematic Diagram of Experimental Assembly

When the efficiency of generation is independent of the current input there will be a linear relationship between sample concentration and titration endpoint. It is usually possible to calculate the unknown sample concentration from measured titration time, but in most cases it is easier to calibrate the system using standard solutions.

For the reaction S + R ⟶ Product, where the sample concentration is M moles/litre and the solution flow rate is f mls/min:

$$\text{The number of moles passing the electrodes per second} = \frac{fM}{6 \times 10^4}$$

At the equivalence point of the titration the current applied to the electrodes must generate the same number of moles per second. With a single electron oxidation or reduction (n = 1) and assuming 100% current efficiency the equivalence point will be reached when the current ramp reaches : n.f.M.F. / 6 x 10^4 amps, where F is the Faraday constant.

CHAPTER 18

The Automated Analysis of Cyanide in the Presence of Sulphide by Gradient Titration

18.1. Experimental

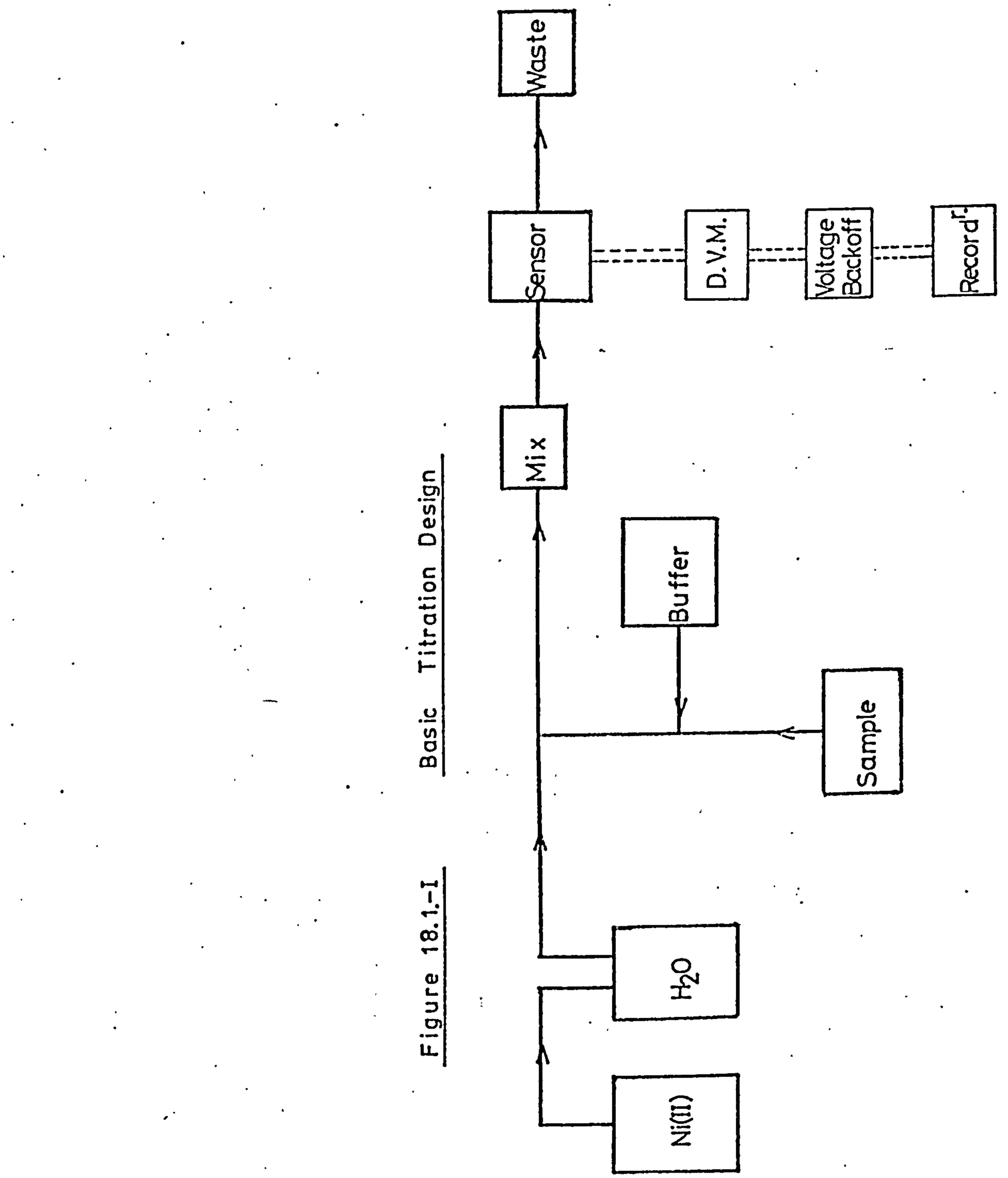
18.2. Results and Discussion

18.3. Conclusions

Cyanide and sulphide frequently occur in the same sample, and although sulphide is a serious interferent for the cyanide electrode it has been shown that the effects of sulphide can be masked out to allow the measurement of cyanide activity with a silver iodide based ion selective electrode. In this work the continuous analysis of cyanide in the presence of sulphide has been attempted, by titration with nickel using the two cup gradient titration technique. The sulphide in solution does not change the amount of nickel added to reach equivalence, but it does decrease the potential change during the titration thus preventing the analysis of low cyanide levels in the presence of excess sulphide. The effects of masking by cadmium in a flow stream were investigated in an attempt to produce a measurement system for the automated titration of cyanide in the presence of sulphide.

18.1. Experimental

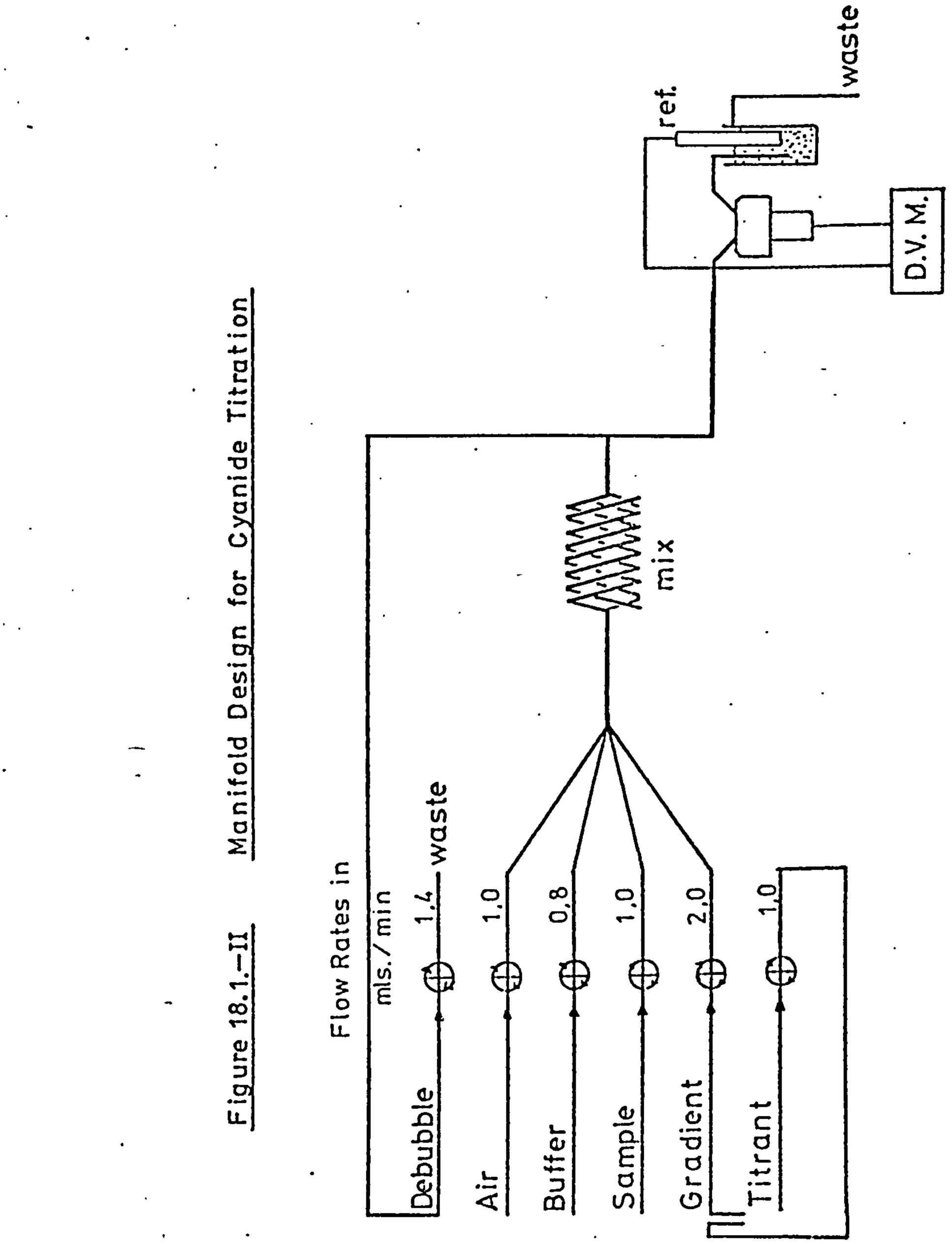
The two cup gradient titration analysis was carried out using the basic experimental design shown in Figure 18.1.-I with the manifold for the cyanide titration shown in Figure 18.1.-II. All tubings were



Basic Titration Design

Figure 18.1.-I

Figure 18.1.–II Manifold Design for Cyanide Titration



calibrated to ensure a linear concentration gradient and the solutions were pumped by a Technicon Auto Analyzer proportioning pump with air bubble segmentation to aid reagent mixing and suppress pulsing.

Nickel sulphate was used as the titrant and an initial volume of distilled water (V_{Bo}) of 10 mls was placed in vessel II to give a titration time (D) of 10 minutes. The buffer used for non-masked titrations was 0.5M NH_3/NH_4Cl, and 0.5M NH_3/ NH_4Cl, 0.02M $CdCl_2$ for the masked titrations.

A silver iodide/silver sulphide (1:1) membrane with direct contact and flow through cap was used as the endpoint sensor, with a S.C.E. reference electrode placed downstream.

The reproducibility of ion-selective electrode endpoint monitoring was evaluated and the sensitivity of the titration technique for cyanide samples was determined.

18.2. Results and Discussion

A typical potential/time plot of a titration is shown in Figure 18.2.-I. To avoid any uncertainty in establishing when the first segment of gradient reaches the sensor, the dead time is included in the titration time. This is not strictly necessary but when a titration time is used in conjunction with a calibration graph the sample concentration may be found by interpolation, or the dead time calculated from the intercept of the time axis. The indicated potentiometric endpoint was found by extrapolation of the curve (Figure 18.2.-II).

Figure 18.2.-I

A Typical Potential vs. Time Plot of a Titration

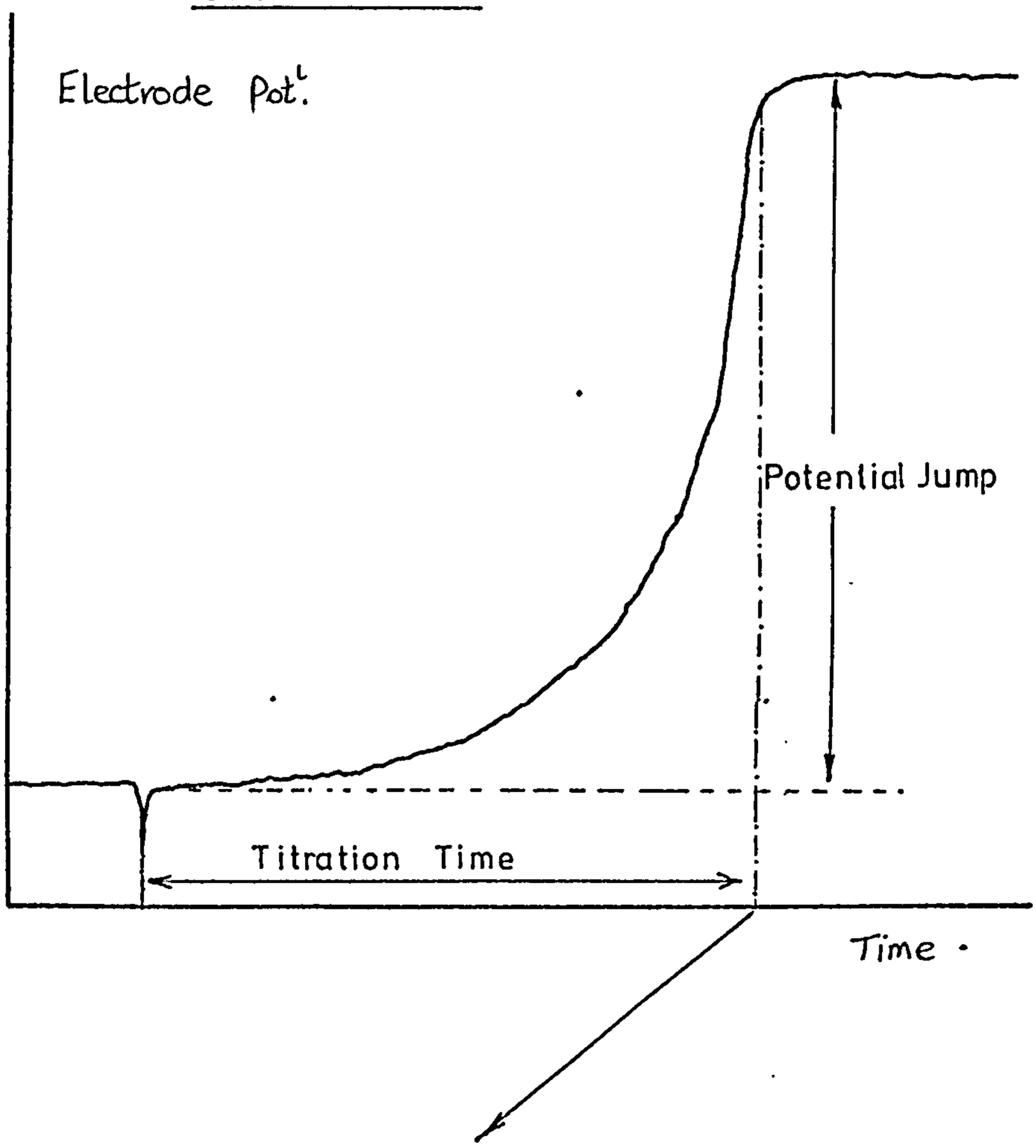
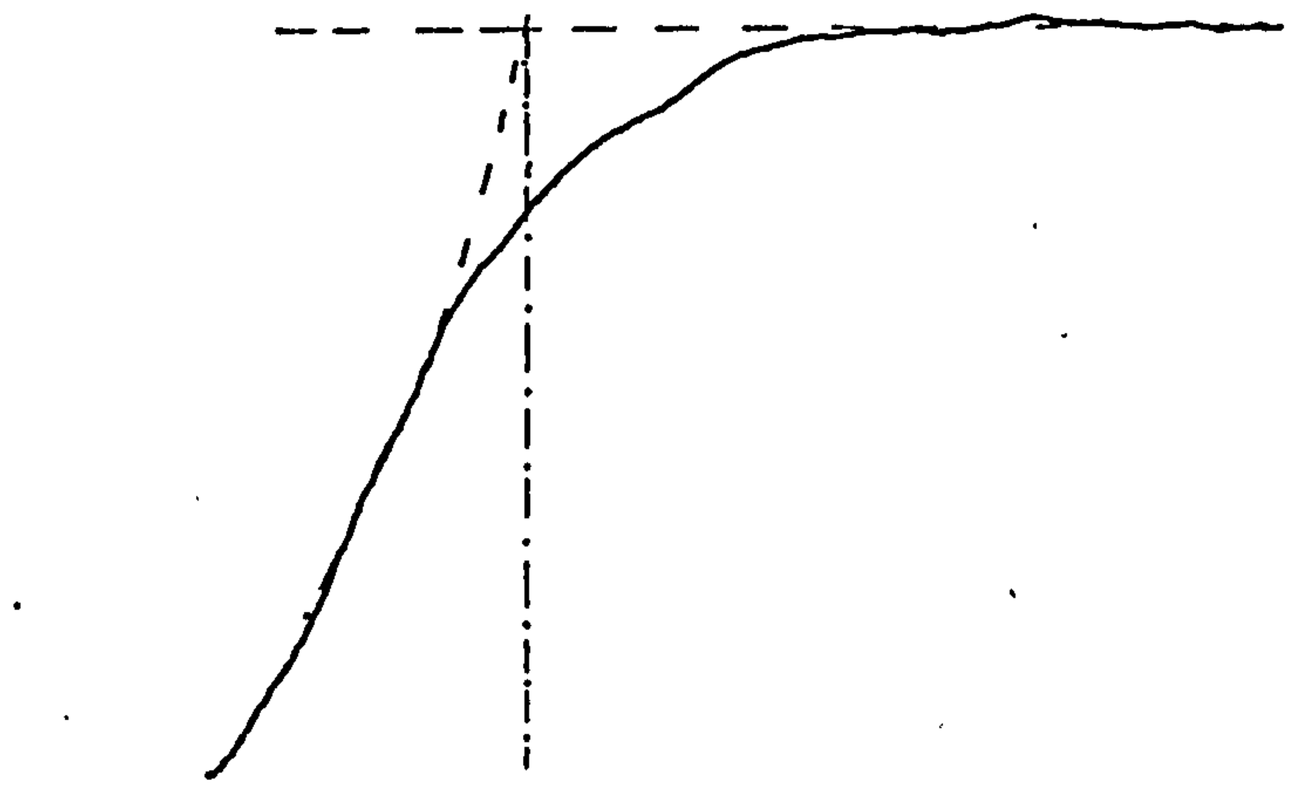


Figure 18.2.-II Endpoint Extrapolation



The reproducibility of the method was tested by titrating several different portions of a potassium cyanide sample with a nickel titrant, the results for which are given in Table 18.2.-III.

Table 18.2.-III Reproducibility of Gradient Titration Of Cyanide

Sample 10^{-3} M KCN. Titrant 5 x 10^{-4}M $NiSO_4$

F_A = 1.0 ml/min, F_B = 2.0 ml/min, V_B = 10.0 mls

Titration Time (mins)

2.33, 2.20, 2.45, 2.35, 2.32
2.33, 2.35, 2.32, 2.30, 2.45

Mean = 2.34 mins (Theoretical = 2.5 mins)

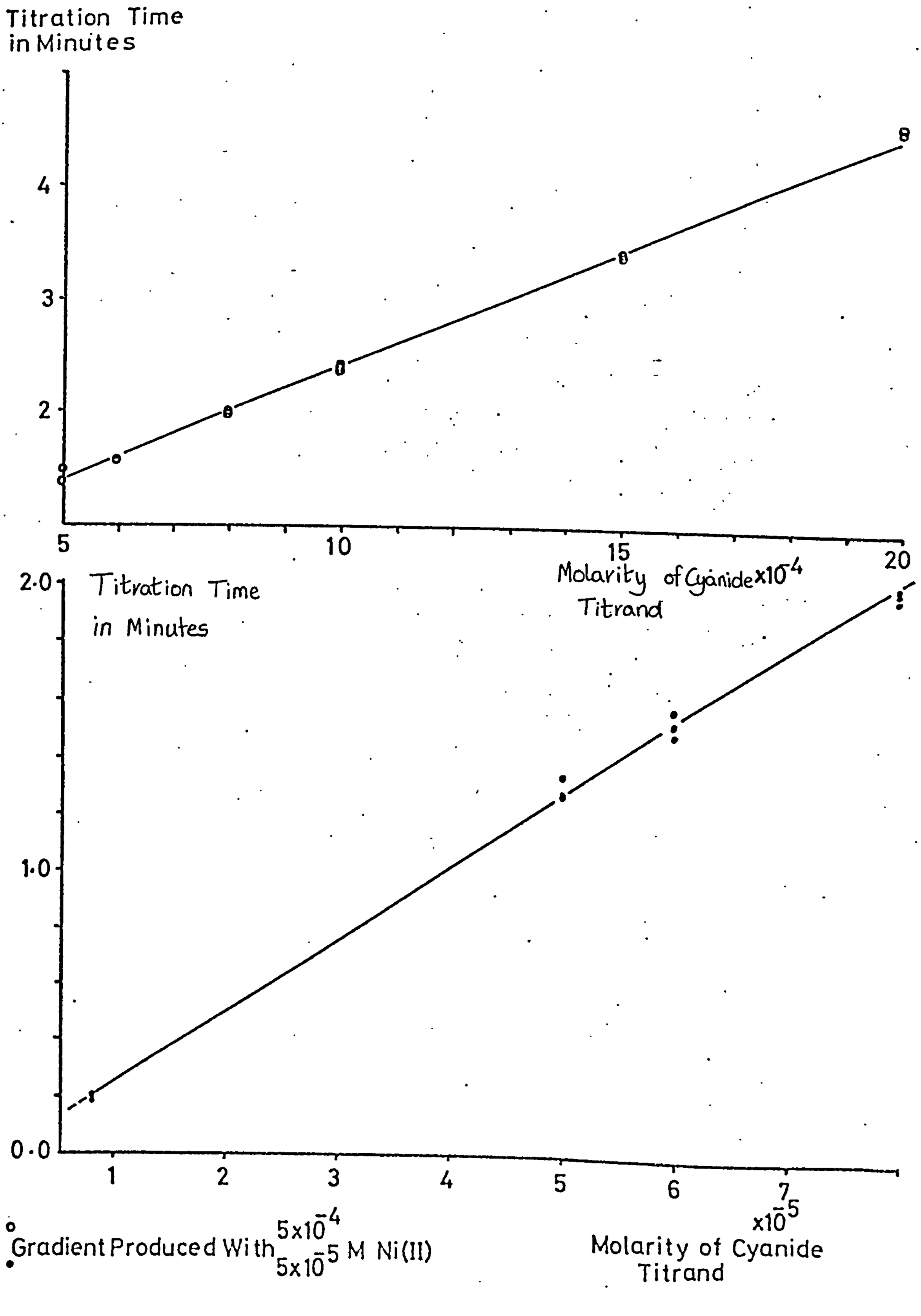
Standard Deviation = 0.072

Relative Standard Deviation = 3%

As the titration time is reasonably reproducible but not the same as the theoretical value, probably due to a slightly non linear gradient, several different concentrations of cyanide were analysed to obtain a calibration graph of titration time *vs.* sample concentration. The results shown in Figure 18.2.-IV, indicate a linear relationship between concentration and titration time for cyanide in the range of 8 x 10^{-6} M to 2 x 10^{-3} M. As the concentration of cyanide is decreased the potential difference between the starting point and endpoint also decreases, for example the change in potential during the titration of 10^{-3} M CN^- is approximately 50 mV, whereas for the 8 x 10^{-6} M solution it is reduced to only 3 mV. The cyanide electrode, in a flow stream, can easily give a noise level of $\pm$ 0.5 mV and thus the 3 mV difference in

Figure 18.2.-IV

Calibration Graph of Cyanide Titration Results



potential at an endpoint obtained from the 8×10^{-6} M CN^- in pure solution is considered the detection limit by gradient titration.

The effects of sulphide on the analysis were determined by adding different amounts of sulphide ion to a cyanide solution, and titrating the mixture. The potential/time curves for the titrations are shown in Figure 18.2.-V, and the results are given in Table 18.2.-VI.

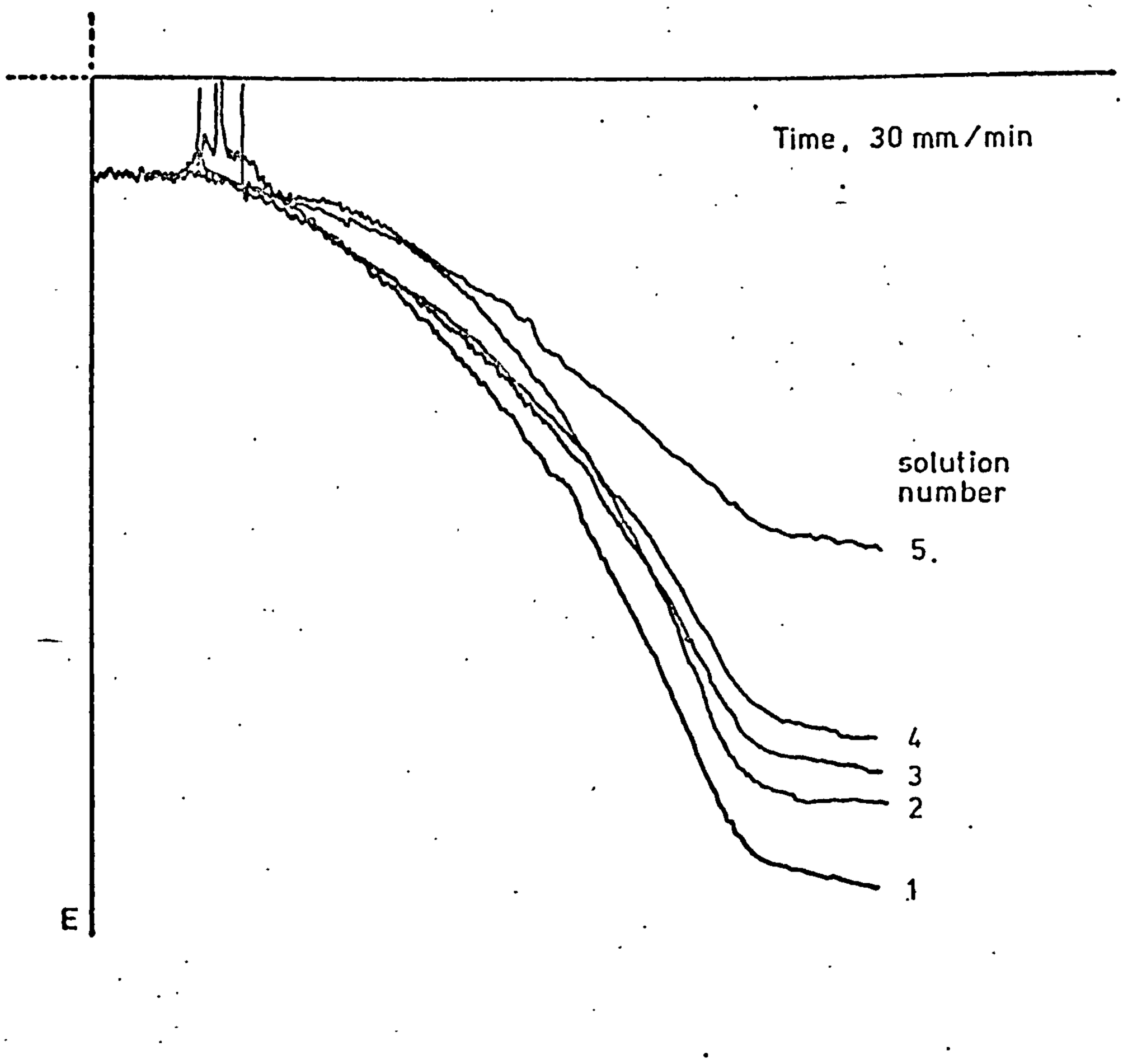
Table 18.2.-VI Results of the Cyanide/Sulphide Mixed Solution Titrations

Solution	$[S^{2-}] \times 10^3$M	$[CN^-] \times 10^3$M	Titn.Time (mins)
1	0	1	2.35
	"	"	2.34
2	0.25	"	2.34
	"	"	2.33
3	0.50	"	2.33
	"	"	2.32
4	0.75	"	2.34
	"	"	2.47
5	1.0	"	2.38

In the nickel titration one mole of sulphide (NiS) is equivalent to four moles of cyanide ($Ni(CN)_4^{2-}$) but the effects of the ion on the titration time are not apparent until its concentration is almost equivalent to that of the cyanide. The potential/time plots do show the effects of increasing sulphide activity. During the titration the sensor monitors the change in activity of the mixed solution until, at the equivalence point, it responds only to the sulphide ion remaining in solution. Thus an increased concentration of sulphide interferent will cause an associated decrease in the potential change at the endpoint.

Figure 18.2.-V

Potential vs. Time Curves for the Cyanide/Sulphide Mixed Solution Titrations



The results for the cadmium masked titrations are given in Table 18.2-VII. The titration times were the same for cyanide plus masked sulphide as they were for cyanide

Table 18.2.-VII. Titrations of Cyanide/Sulphide Mixed Solutions With Cadmium Masking - Potential Jump at Endpoint

Conc. CN^- (Molar)	$4x10^{-4}$	$8x10^{-4}$	$10x10^{-4}$	$12x10^{-4}$
Conc. S^{2-} (Molar)	Potential Jump at Endpoint (mV)			
0	6	13	28	29
$4x10^{-4}$	1	8	21	22
$8x10^{-4}$	*	6	14	14
$12x10^{-4}$	*	4	7	10
$16x10^{-4}$	*	*	*	7

Cd Cl_2 masking at $3x10^{-3}$ Molar, * no endpoint visible

alone at the same concentration, upto the point where the endpoint became indistinguishable.

18.3. Conclusions

An ion selective electrode can be used as a sensor in the automated analysis of cyanide to a lower level of $8x10^{-6}$ Molar. With cadmium masking, in an automated system of cyanide analysis, a sulphide level almost twice that of the cyanide can be tolerated before the endpoint becomes indistinct. In this study the endpoint could be observed when the potential jump was twice the size of the signal noise (usually $\pm$ 1mV) and the accuracy of the endpoint was maintained up to the same sulphide concentration. When cadmium was not added, an

error was introduced into the titration time before the endpoint became indistinct. The poor masking effect is probably due to the fact that in a flow stream the cadmium sulphide is passed over the surface of the silver iodide membrane,allowing exchange of the sulphide between the cadmium and the silver. This is indicated by a soft black layer which forms on the membrane surface during titrations.

Only one concentration of cadmium was used in this study, and more work should be carried out to investigate the effect of masking agent concentration. As the intention of this work was to devise a method of cyanide analysis in effluents, the addition of a greater concentration of cadmium would cause a more serious pollution problem than it solved. The use of hydrogen peroxide has been suggested for removal of sulphide from solution and could be effective in this application, if it can be shown to have no interference on the cyanide electrode response.

CHAPTER 19

A Fully Automated, Single-Cup Gradient Titration of Chloride, To A Differential Potentiometric Endpoint

19.1. Experimental

19.2. Results and Discussion

19.2.1. Analysis of Aqueous Samples

19.2.2. Analysis of Serum Samples

19.3. Conclusions

The titration of silver ions with chloride is a well known reaction, serving as a good example of the single cup gradient titration technique, for which selective electrodes are available sensitive to both the titrant and the titrand. The automated system could also have some relevance in clinical chemistry as a common routine in laboratories is the determination of chloride content of different body fluids, especially serum. The concentrations of total and bound chloride in the system change when the body is not in its 'normal' condition, varying between 85 and 112 m.eq per litre (153), and can be used as a diagnostic test.

Colourimetric methods are most frequently used for the analysis of chloride (154,155,156) with an accuracy of $\pm$ 2% on titrations of 50μl samples. Electrochemical techniques have the advantages of speed and accuracy over colourimetric methods and can also be used for the measurement of halides at very low concentrations, by potentiometry at finite current (157,158). The ionic activity of chloride in serum has been studied by direct

potentiometry using ion selective electrodes (153), but the accuracy was not sufficient,possibly due to the variations in potential of a reference electrode in solutions containing particulate matter.

Two selective membranes, sensitive to different ions can be used as a sensing and reference electrode (101, 159) and such a system has been used for the titration of cyanide with silver (160). This latter variant has been adapted to the measurement of chloride by titration with silver, to a silver chloride and silver sulphide differential electrode endpoint.

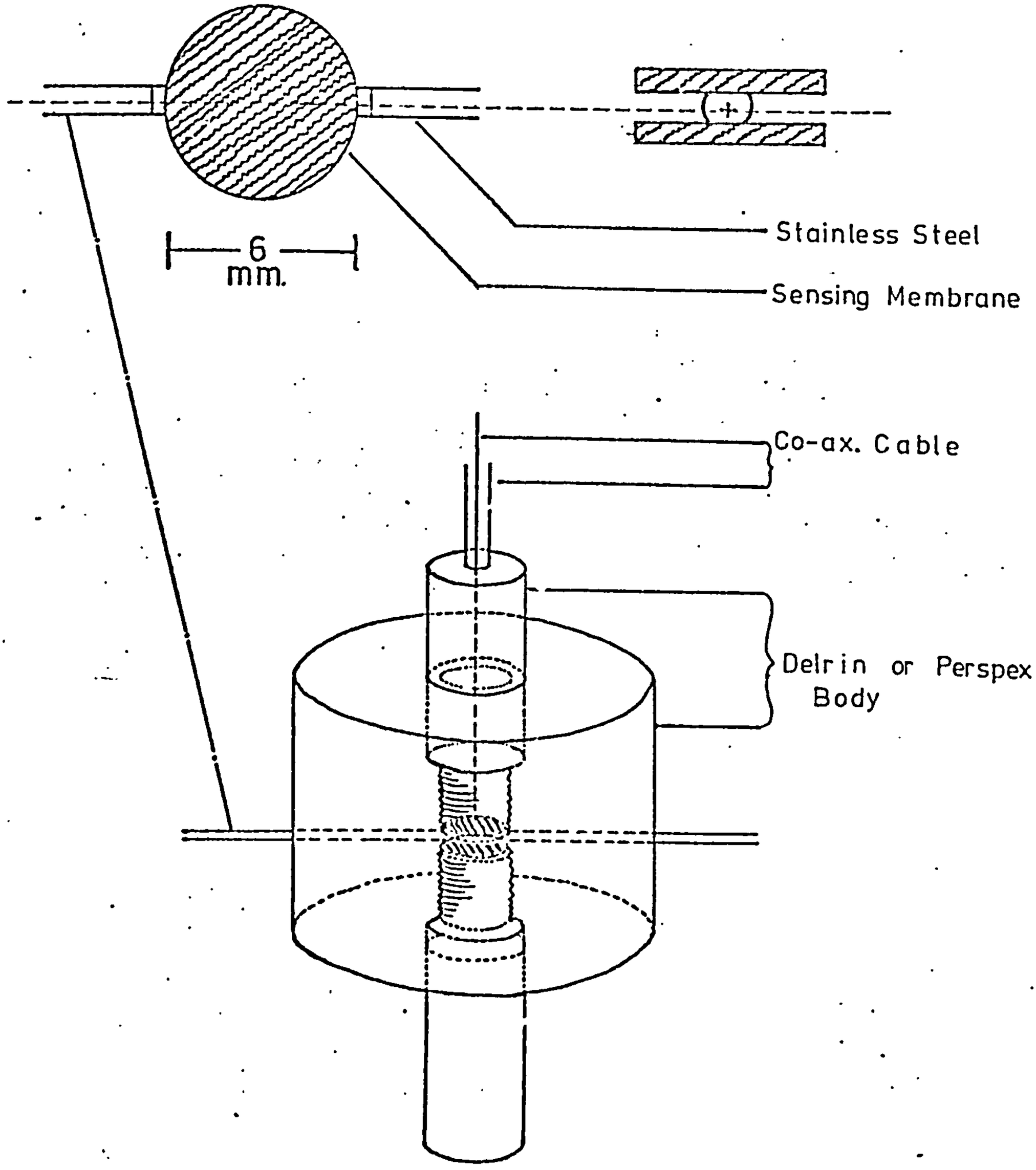
19.1. Experimental

The differential amplifier system used was an Analog Devices instrumentation amplifier (Model 603K) with an impedance of 10^{12} ohms at both inputs to allow the use of two selective membrane electrodes.

The flow cell was designed (Figure 19.1.-I) to have a minimum dead volume and to enable the two membranes to sense ions in the same portion of solution simultaneously. It also creates the minimum disturbance of solution flow.

The electrodes used for the differential titration of chloride are the chloride ion- and the sulphide ion-selective electrodes. These were made by cold pressing $AgCl/Ag_2S$ and Ag_2S respectively in a 6 mm diameter die at a pressure of 2 tons. The membranes were fixed into threaded 'Delrin' plastic tubes which were externally threaded to allow assembly into the flow cell. Contact between the membranes and amplifier was made by double

Figure 19.1.-I


6
mm.
Stainless Steel
Sensing Membrane
Co-ax. Cable
Delrin or Perspex
Body

screened cables and a silver loaded epoxy solid contact.

Stock solutions of $AgNO_3$ and KCl were made from analytical reagent grade chemicals. The potassium chloride solution was standardized by manual titration using a potentiometric endpoint. The standard serum samples were provided by Technicon and had a chloride content of 104 m.eq.$l.^{-1}$.

Manipulation of samples was carried out using Technicon Autoanalyser Modules, a Proportioning Pump III and a Sampler II. Pump rates of the calibrated tubings, were F_A = 1 ml/min and F_B = 2 mls/min. Minor adjustments to the flow rates can be achieved by raising or lowering the reagent vessel.

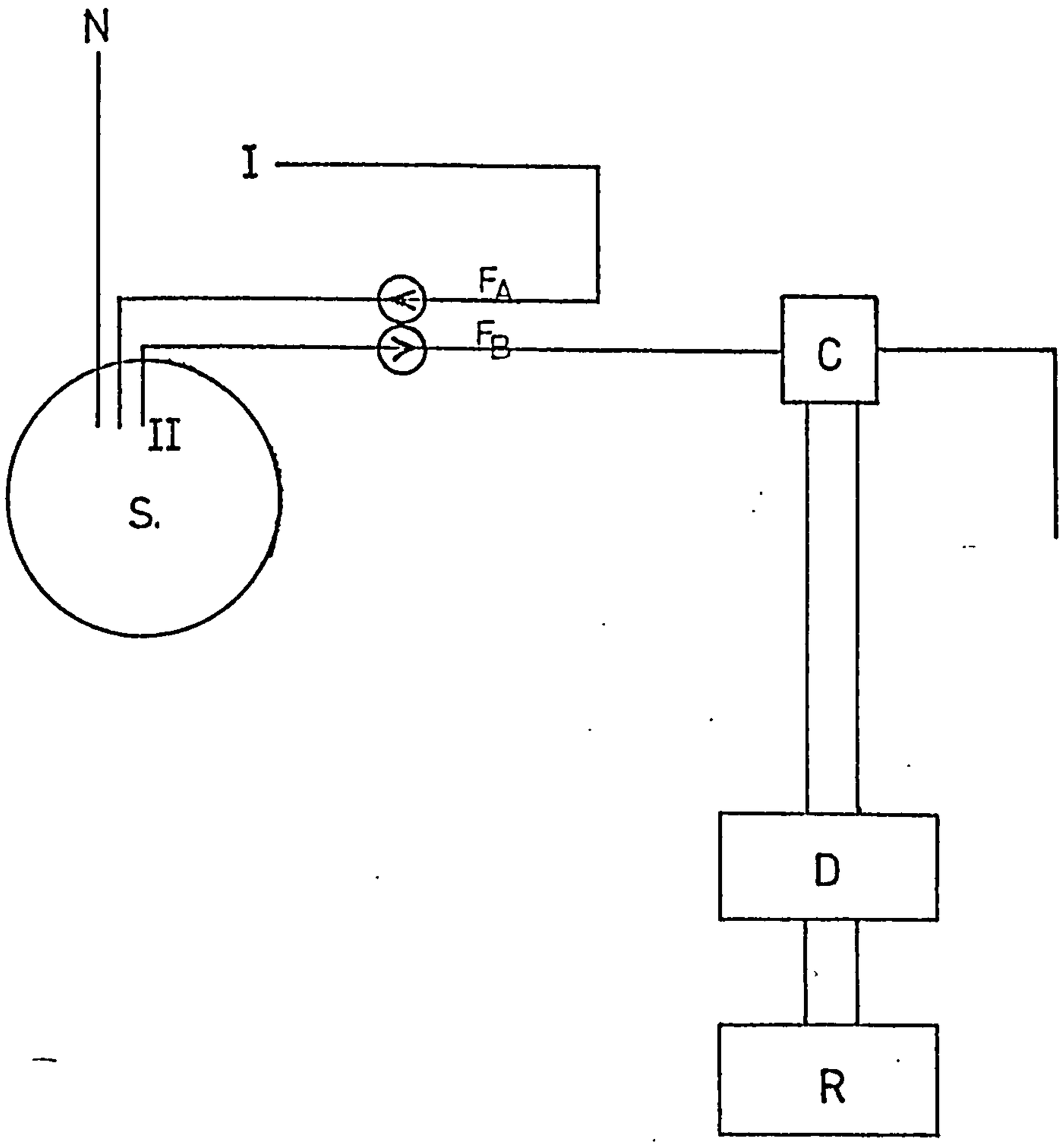
One design of the manifold is shown in Figure 19.1.-II. Reagent from reservoir I is pumped into the sample vessel II (5 ml AutoAnalyser sampler cup) while a rapid stream of nitrogen (N) is passed through to effect complete mixing.

The solution from II is then pumped onwards into the measuring cell (C) and then to waste. It is possible to locate the cell either immediately downstream of the sample cup and prior to the pump or alternatively the pump can be inserted between the cell and sample container. The latter arrangement was used in the present system. The output from the cell is fed to the differential amplifier unit (D) and the potential time profile displayed on a x-t potentiometric recorder (R).

Samples are presented sequentially interspersed by a water wash. The sampling rate is controlled by the titration time which in turn is dependent on the degree of

Figure 19. 1.-II

Titration Manifold Design



N	Nitrogen
S	Technicon Sample Turntable
C	Measuring Cell
D	Differential Amplifier
R	Recorder
⊛	Pump

precision required. The present system operates at a rate of 12 samples/hr. but could be increased significantly by using a more rapid generation of gradient.

Preparation of the serum samples for analysis presented two problems. i) The volume of sample available is small, although the chloride level is high and amenable to extensive dilution. ii) There is a large concentration of protein in the sample which will be strongly adsorbed onto the membrane surface if it is not treated. These problems were solved by adding 0.1 ml of concentrated nitric acid to 125 μl of sample before diluting to 5 ml with distilled water.

19.2. Results and Discussion

19.2.1. Analysis of Aqueous Samples

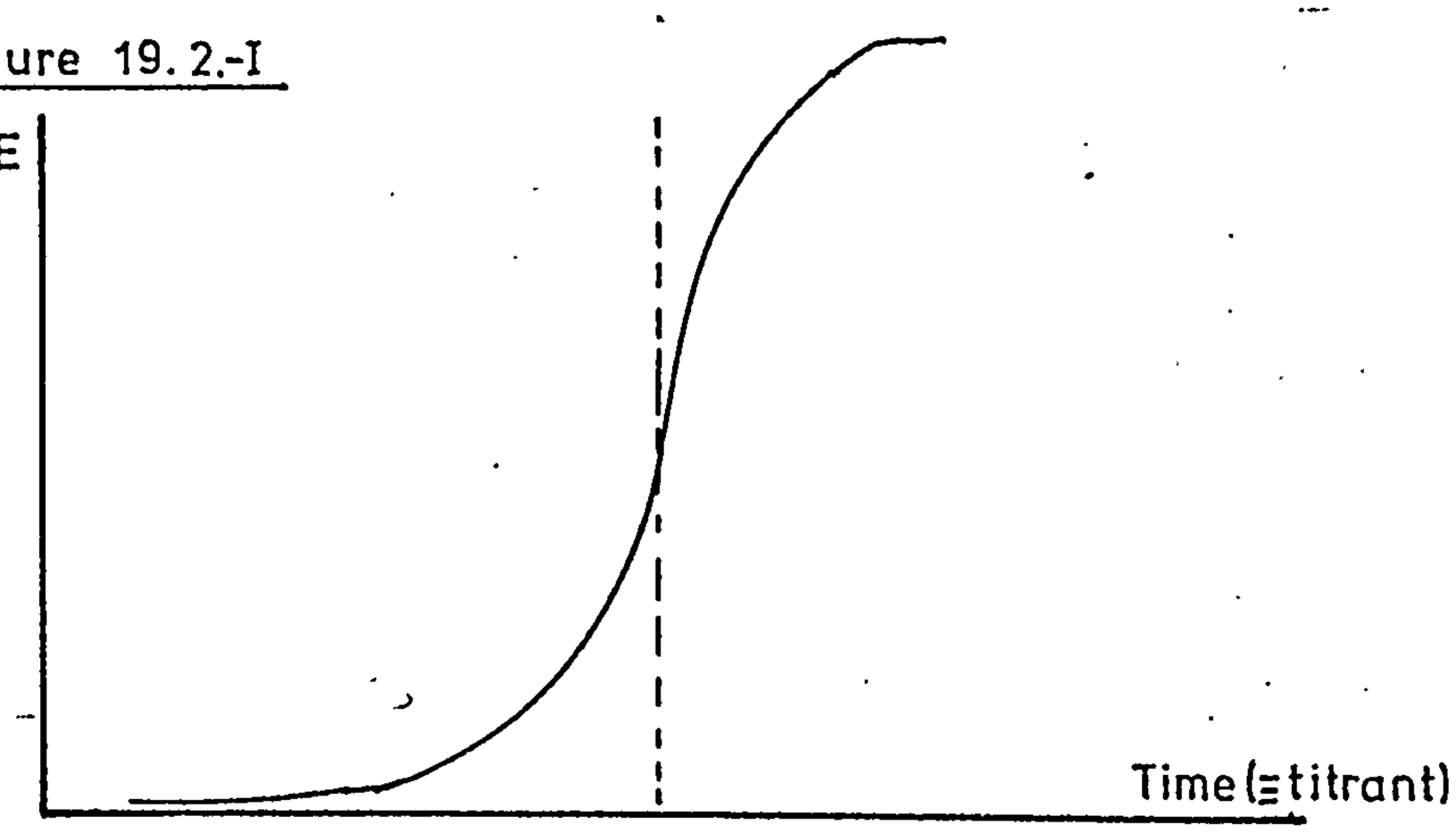
In the preliminary investigation, individual samples of potassium chloride (10^{-3}M) were titrated to a potentiometric endpoint using a calomel reference electrode and either a chloride sensitive or silver sensitive electrode as the indicating system. In both cases the titration curve was the normal 'S' shape common to many potentiometric titrations (Figure 19.2.-I).

The same chloride sample was then titrated using the silver chloride and silver sulphide electrodes as the indicating system to produce a titration curve as shown in Figure 19.2.-II.

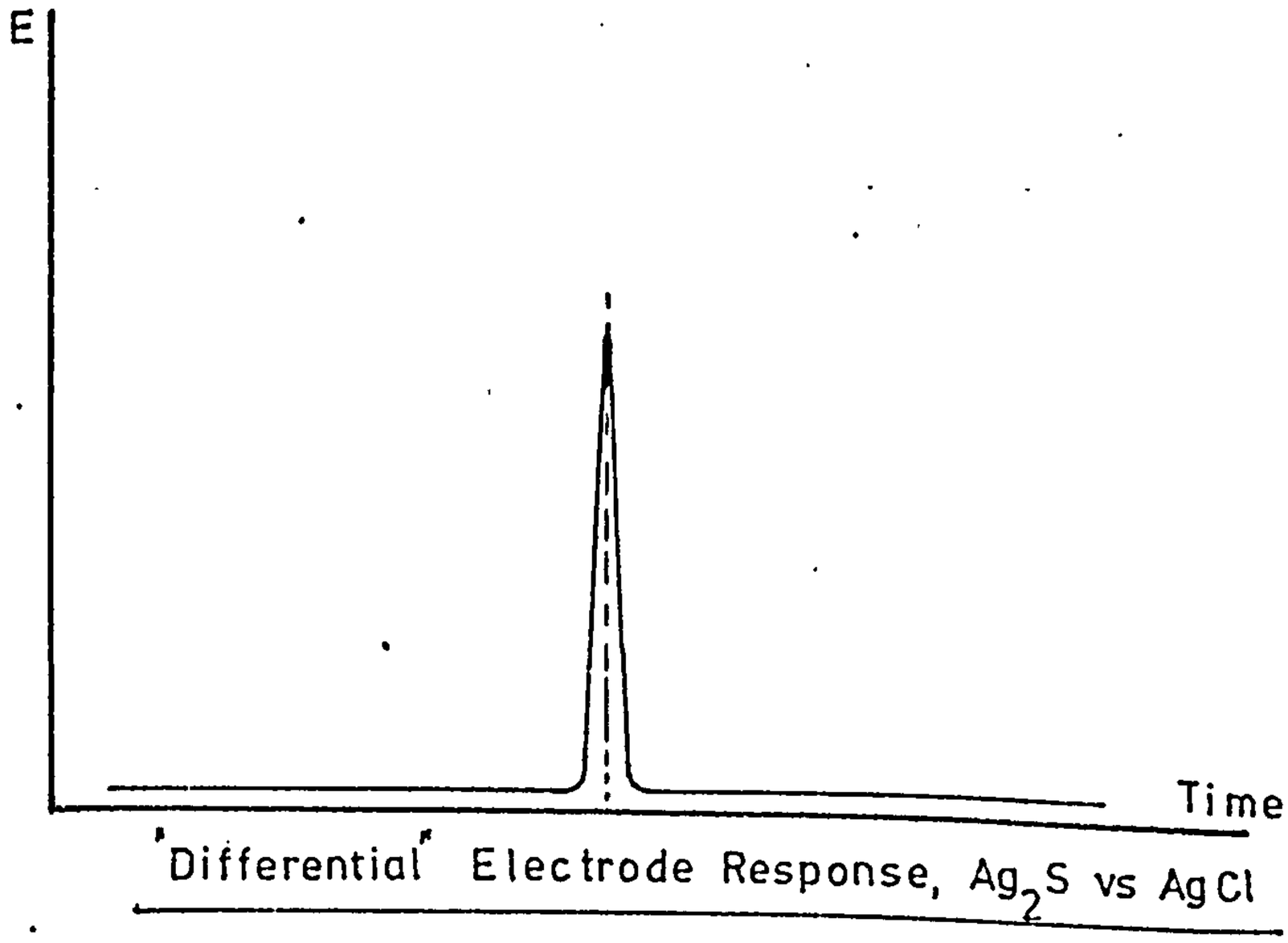
In order to interpret the shape of the curves it is necessary to define the potential determining species for both electrodes in various regions of the titration process.

Simon et al (66) have carried out a detailed study of the response of solid state membrane electrodes and have interpreted the selectivity and detection limit for the silver halide membranes in terms of the equilibrium activity of Ag^+ at the membrane surface. This silver ion activity can result either from the finite solubility of the membrane, or from the intrinsic defect level of Ag^+ within the membrane itself. These principles have been employed in an attempt to interpret the shape of the titration curves in the present system.

Figure 19.2.-I



Membrane Electrode Response vs. SCE



"Differential" Electrode Response, Ag_2S vs AgCl

In the initial part of the titration both electrodes effectively respond to chloride ion; in the case of the sulphide membrane via an interference effect where the potential can be expressed as

$$E = E^{o} - \frac{RT}{F} \ln (K_{S/Cl} \, a_{Cl^-})$$

In the titration region close to the endpoint, the potential of the chloride membrane becomes constant since the solubility of the membrane controls the activity of Ag^+, which is very much greater than the defect level of Ag^+. The predicted electrode potential changes are shown schematically in Figure 19.2.-III. The potentials of both electrodes will initially follow the decrease in chloride concentration until the effective p Ag^+ is in the region of 5.5 which represents the detection limit for chloride ion. In this region the potentials of both electrodes remain almost constant. The point is also defined by the defect activity level of Ag^+ in both membranes. With increasing additions of Ag^+ titrant, the sulphide membrane will be the first to show a response due to its lower detection limit for Ag^+. The differing responses of the two membranes in this region cause the peak shaped response of the electrode system. With further addition of titrant, the Ag^+ level will eventually exceed the detection limit of the silver chloride membrane, defined by the solubility product of AgCl and subsequently both electrodes will exhibit a similar response to further increase of silver ion. This interpretation is further supported by the response obtained for a silver iodide, silver sulphide electrode pair.

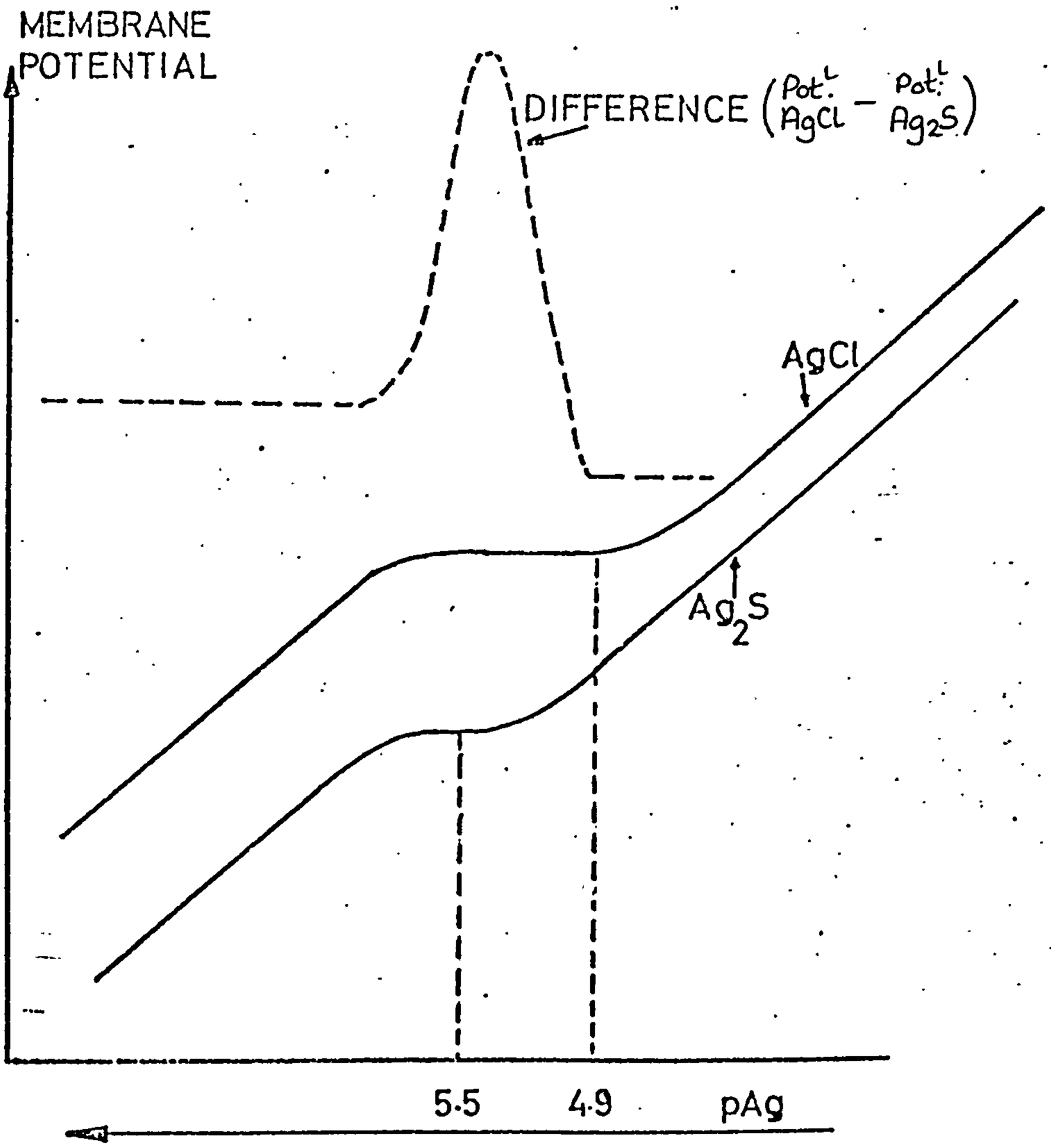


Figure 19.2.–III

Predicted Electrode Potential Changes

In this case, where the detection limit of both electrodes is controlled by the defect level of Ag^+, no differential peak response was observed (160).

To investigate the validity of using the peak as the endpoint, a series of chloride solutions were prepared, titrated, and the observed endpoints (peaks) were compared with the calculated ones (Table 19.2.-IV). Typical titration curves for the automated system and sampler sequencing are shown in Figure 19.2.-V. A graphical plot of observed *vs.* calculated endpoint (Figure 19.2.-VI) and the calibration graph (Figure 19.2.-VII) deviate from theoretical at the higher concentration levels. This is due to a slightly non-linear gradient caused by variation in the pump rates, but would not invalidate the use of the peak endpoint if the calibration graph was used.

19.2.2. Analysis of Serum Samples

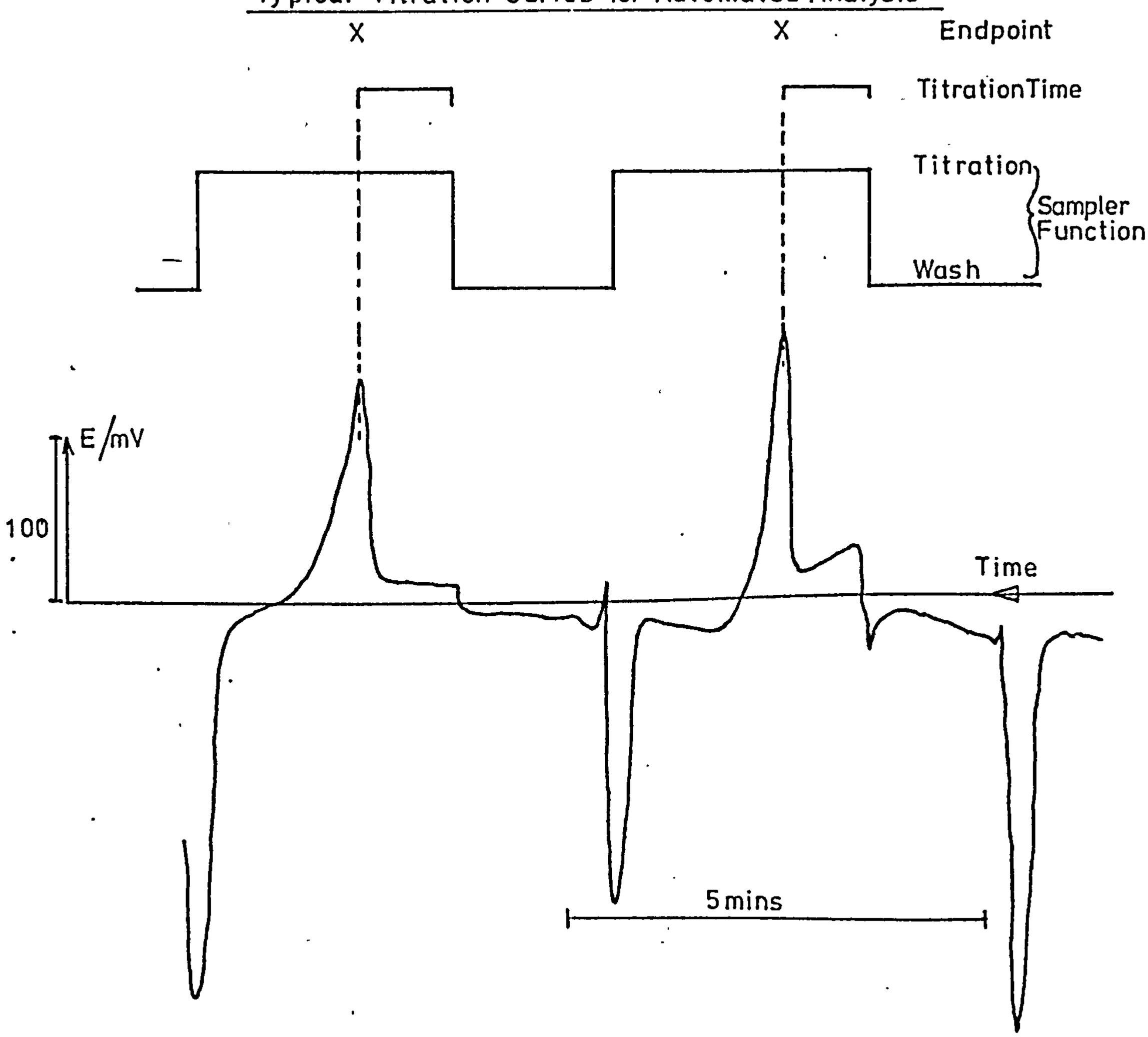
The titration curves obtained for serum chloride determination (Figure 19.2.-VIII) are slightly less well defined than those for pure aqueous standards but, nevertheless, the endpoints are readily identifiable. The change in the shape of the curve was shown to be caused by the addition of acid to the solution, as standards prepared for the calibration graph in the same way had the same shape. The effect was probably due to the changes in solubility of the silver chloride, present in both product and sensing membrane. The results for both the standard serum and calibration solutions are given in Table 19.2.-IX.

Table 19.2.-IV Results of Chloride Analysis In Aqueous Samples

Concn. Cl^- x 10^{-3} M	Calculated Endpoint (3.0=60 sec)	Observed Endpoint	Mean
1	2.5	3.0,2.9,3.0	2.97
2	4.38	4.8,4.6,4.6	4.67
3	5.65	6.05,6.1,6.0	6.05
4	6.65	7.15,7.0,7.0	7.05
5	7.50	8.15,8.2,8.4	8.23
6	8.2	9.7,9.4,9.4	9.50
7	8.8	9.9,9.9,9.5	9.80
8	9.25	10.4,10.5,10.1	10.33
9	9.7	10.8,10.6,11.0	10.8
10	10.05	11.2,11.3,11.25	11.25

Figure 19.2.-V

Typical Titration Curves for Automated Analysis



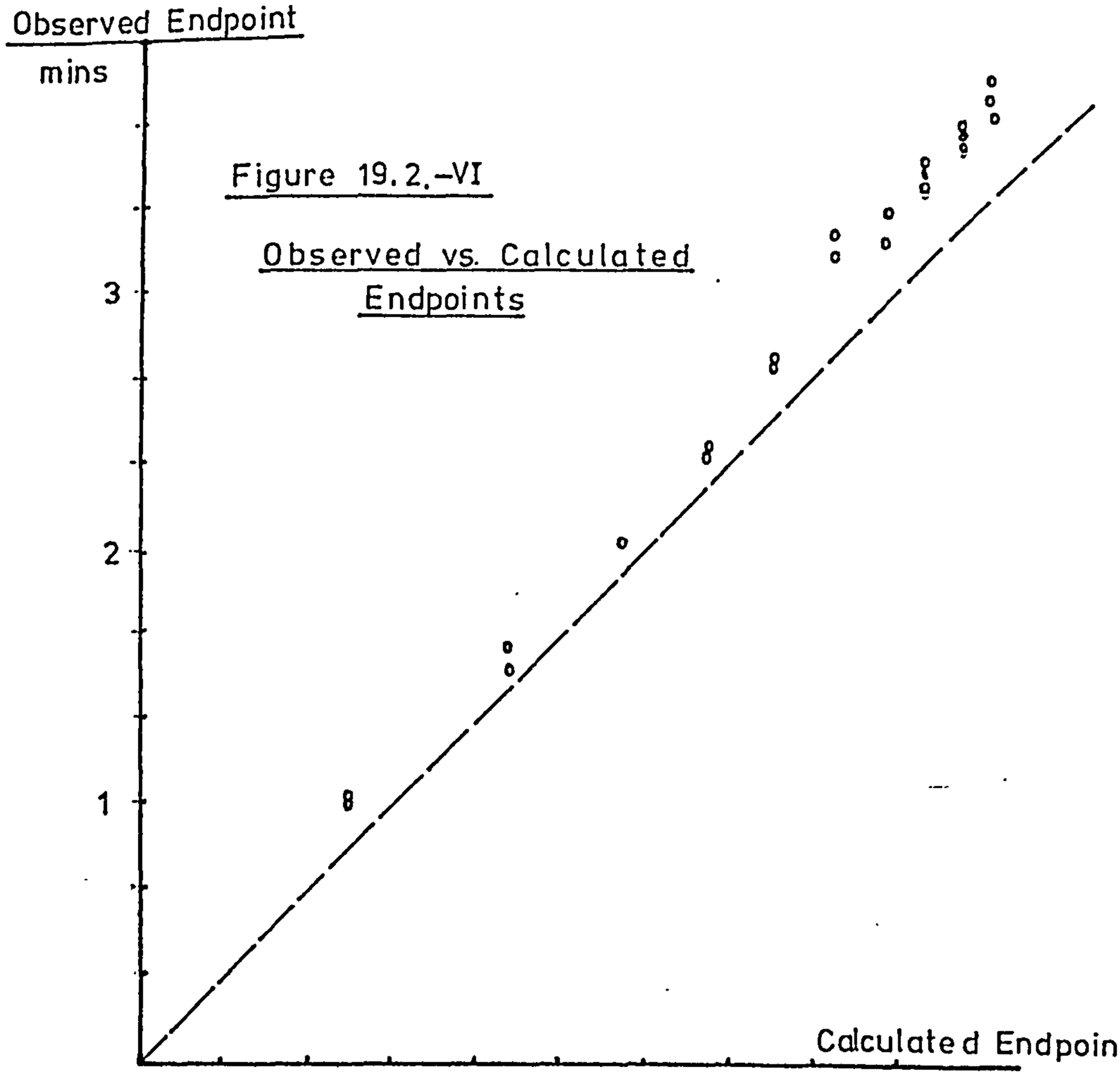


Figure 19.2.-VI

Observed vs. Calculated Endpoints

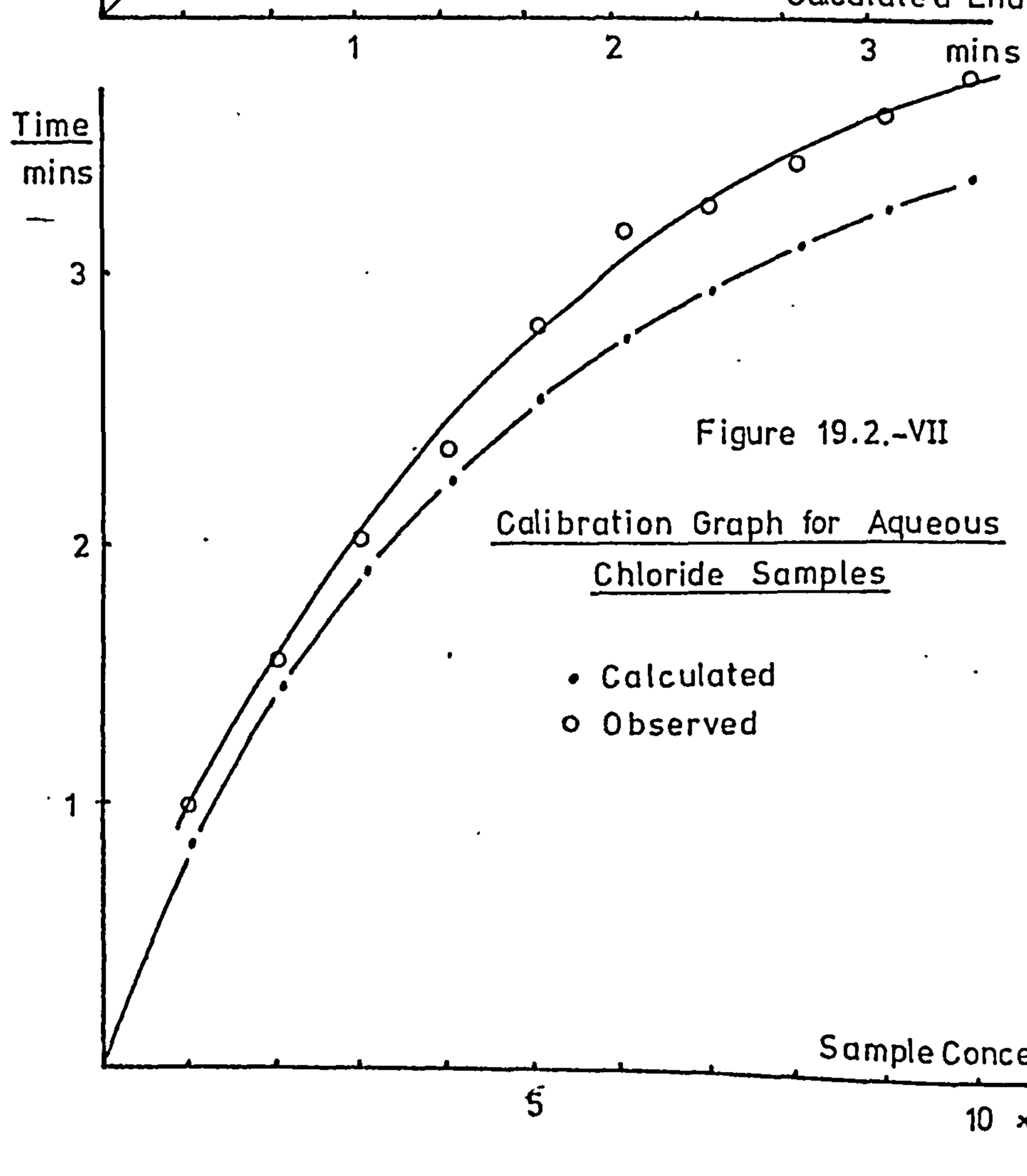


Figure 19.2.-VII

Calibration Graph for Aqueous Chloride Samples

Figure 19.2.-VIII

Titration Curve for Serum Chloride Determination

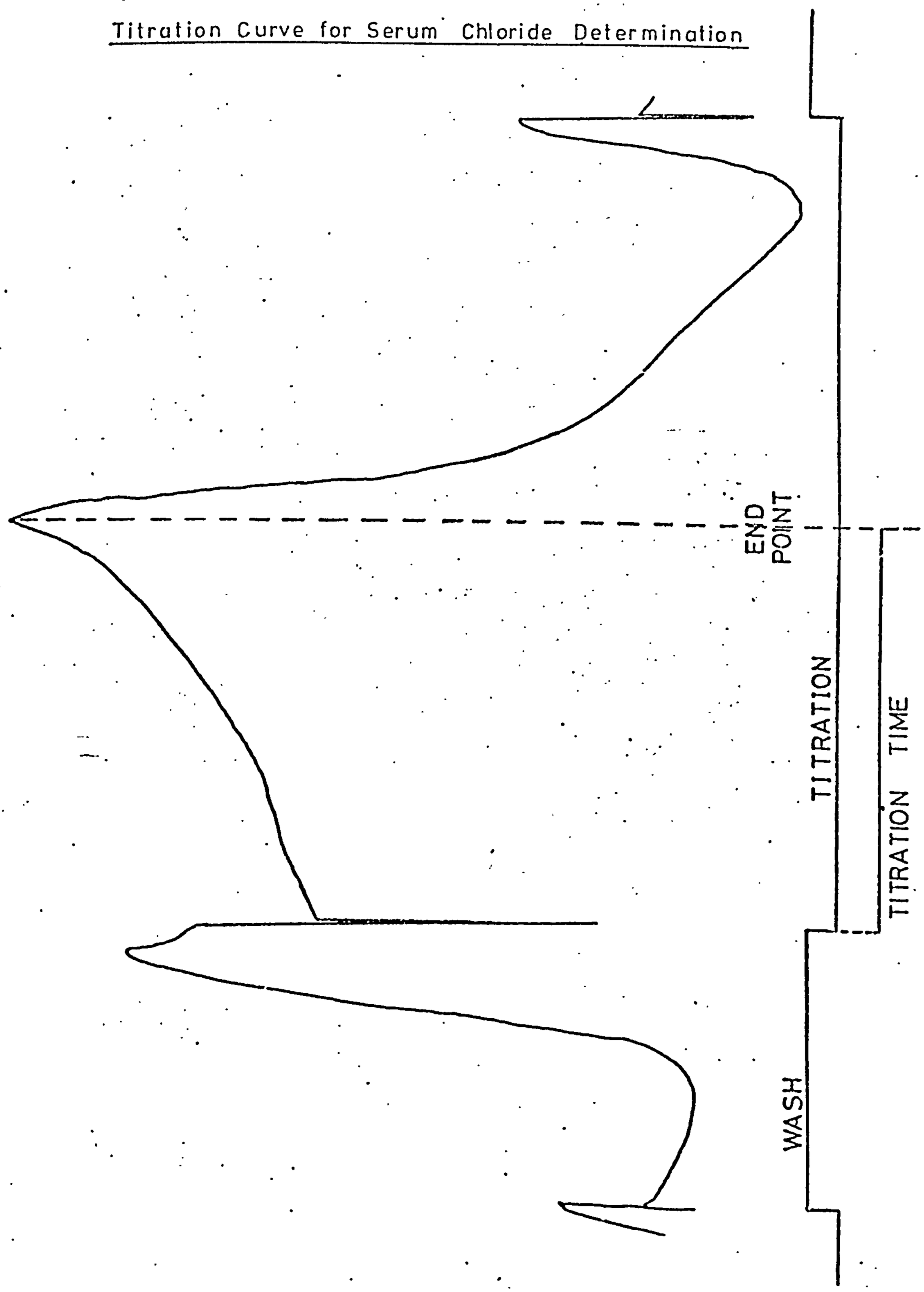


Table 19.2.-IX Results For Serum Analysis

Titration Times For Standard Serum: 6.70,6.70,6.70,6.70,6.67,6.9, 6.75,6.75,6.4,6.85

Mean = 6.71
Std. Deviation = 0.13
Rel. Std. Deviation = 1.97%

Stds. Conc x 10^3 M.Cl-	1	2	3	4	5
Endpoint	4.23	5.91	7.15	8.12	9.0

By interpolation Cl^- = 2.615×10^{-3} Molar

$$Cl^- \text{ in Sample} = \frac{2.615 \times 5 \times 10^{-3}}{0.125} = \underline{104.6 \text{ m.eq.l}^{-1}}$$

Value Stated By Technicon = $\underline{104 \text{ m.eq.l}^{-1}}$

19.3. Conclusion

The method presents a convenient approach to the automation of a wide range of titration systems and is easily assembled using standard AutoAnalyzer modules. Although the precision of the titration is dependent to some extent on the time required for titration, acceptable results can often be obtained with titration times as short as 10 seconds. This is particularly feasible when electronic timing systems are used. Such a system with a 'peak follower' to identify the endpoint and initiate the change of sample - coupled with an accurate concentration gradient - would provide an automatic titrator within the definition given by I.U.P.A.C.

CHAPTER 20

Application Of The Chloride Selective Electrode To Automated Coulometric Gradient Titrations

20.1. Experimental

20.1.1. Instrumentation

20.1.2. Evaluation

20.1.3. Application

20.2. Results and Discussion

20.2.1. The Current Ramp Generator

20.2.2. Application of the Coulometric Gradient Generator to the Titration of Chloride Ions

20.3. Conclusions

Initial studies on this technique of titration used a two cup system for the concentration gradient generation and recently a simplified system using a single cup has been described. The experimental arrangement can be modified even more, however, and the sample titrated in a single pump tube using a coulometrically generated titrant. Current flow can be measured and regulated easily and thus the system promises more accuracy than can be achieved at present, using normal pumped streams for gradient generation. Due to the high current efficiency of silver generation, titrations of chloride with this ion have been very popular (157,158), and coulometric generation is the basis for a commercial chloride titrator, the Radiometer CMT 10 (161).

The present work describes the design and evaluation of a coulometric gradient titration system and its application to the analysis of chloride to an ion selective electrode endpoint.

20.1. Experimental

20.1.1. Instrumentation

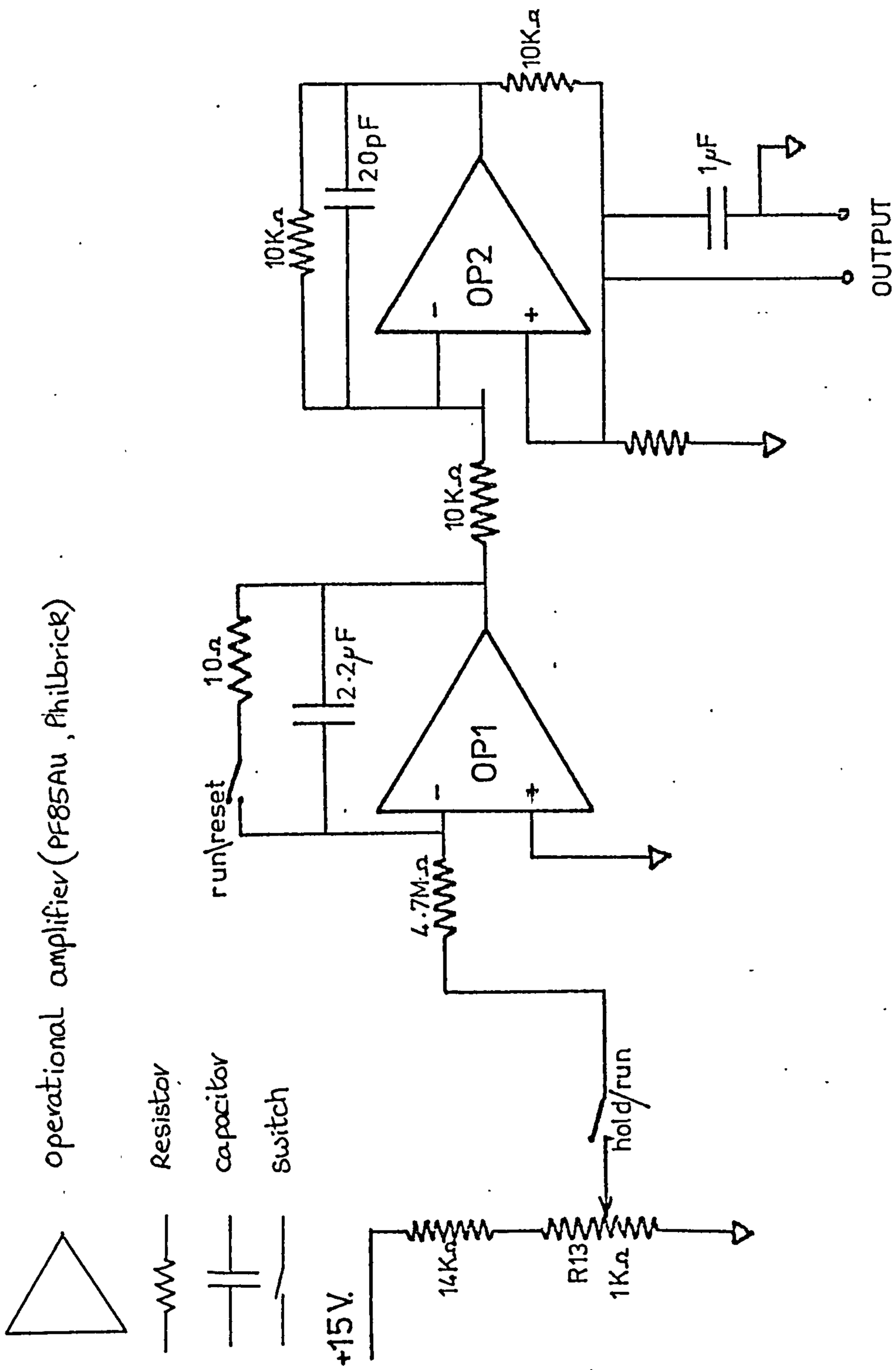
In order to coulometrically generate a concentration gradient in a flow stream, the current flowing between the generating electrodes must vary linearly with time. This can readily be achieved with operational amplifiers and the current generating system for this work consisted of a simple voltage ramp generator and a voltage to current converter. The circuit, shown in Figure 20.1.-I was built, in which the current output is controlled by the voltage input to amplifier OP2, and the voltage ramp slope is adjusted by a ten turn potentiometer R13. The integrator has a time constant of 10 seconds, thus with an input voltage of 1 volt to OP1 the current gradient is 10 μA/second.

The electrodes used for the generation of silver ions were a silver tube (10 mm x 1.5 mm diameter, 45 mm^2 area) located immediately before the measuring cell, and a platinum counter electrode (in 2 molar sulphuric acid) separated from the sample line by a porous glass junction.

Two types of measuring electrode assembly were used. The first approach used the direct measurement mode with either a solid state chloride or sulphide membrane and a saturated calomel reference electrode. The second method used a differential electrode measurement technique where both chloride and sulphide sensing membranes were used together.

A manifold design of the experimental assembly is shown in Figure 20.1.-II. Manipulation of the sample and reagent lines is carried out with AutoAnalyzer modules,

Figure 20.1.-I



The Current Ramp Generator

pump III and Sampler II. In this simple case, a single pump line is used with generation of reagent directly into the sample stream. Typical flow rates in the range of 0.4 to 2.0 mls/min have been used with the present system.

20.1.2. Evaluation

The linearity of the current ramp was initially checked by measuring the voltage drop across a 1 KΩ precision resistor acting as a load. A more realistic evaluation was carried out by measuring the voltage drop across a 100Ω resistor placed in series with the electro-generating cell.

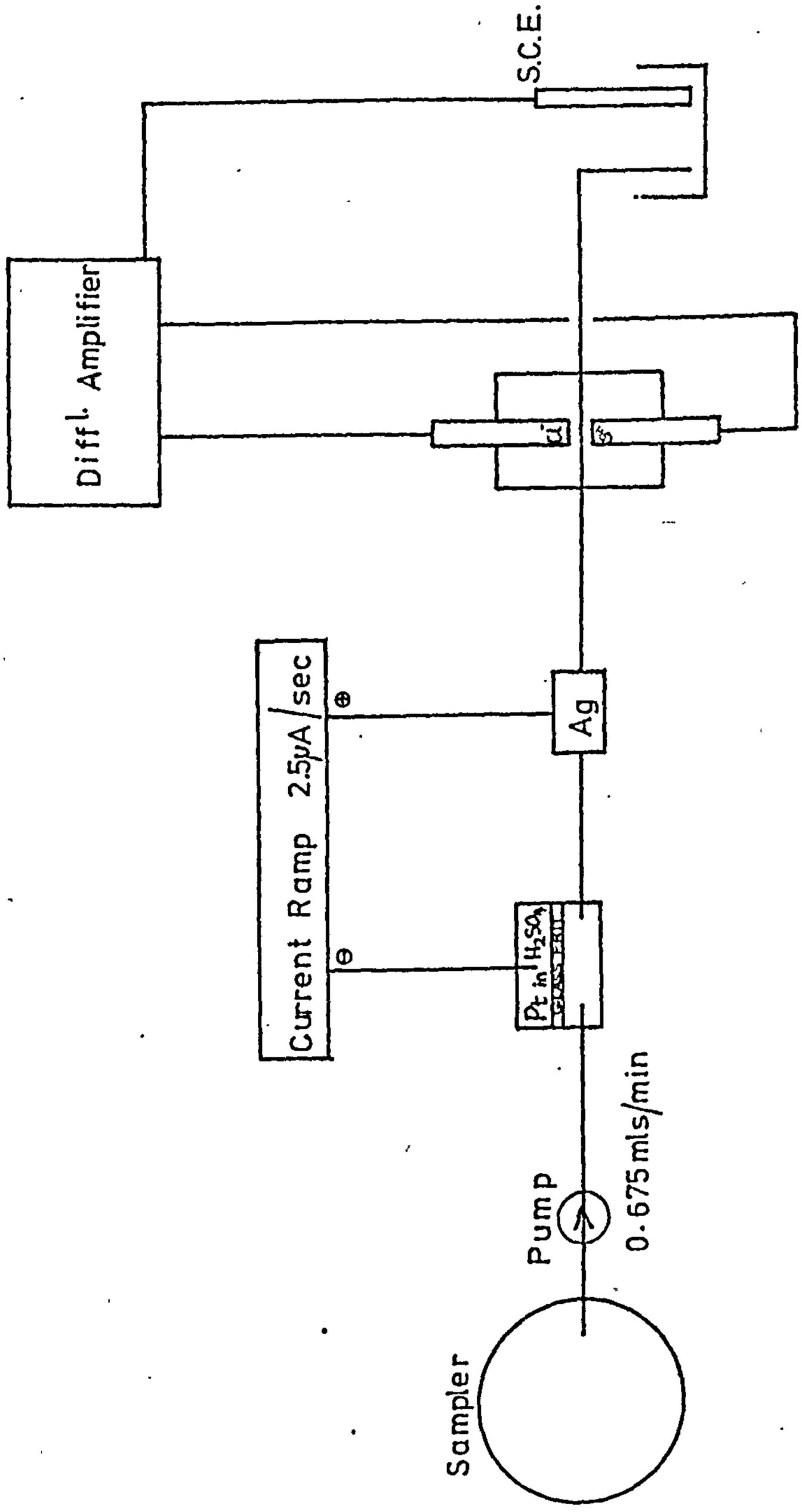
The linearity of silver generation was investigated by operating the current ramp at various gradients in 10^{-1} molar potassium nitrate, measuring the silver activity produced with an ion selective electrode, and plotting the silver activity *vs.* time graph.

20.1.3. Application

The coulometric gradient generator was used in the titration of aqueous samples of chloride ion in the concentration range from 5×10^{-5} to 3×10^{-4} Molar, in a background electrolyte of 0.4 M H_2SO_4 and 0.1% poly(vinyl alcohol). Direct potentiometric and differential potentiometric endpoints were both used for comparison as sensors for automated titrations.

Figure 20.1.–II

Titration Manifold Design



20.2. Results and Discussion

20.2.1. The Current Ramp Generator

The voltage drop across a 1 KΩ resistor, and a 100 ohm resistor in series with the electroactive cell is shown in Figure 20.2.-I. The results indicate that although the generator is capable of providing a high current through a simple resistance, only 0.75 mA could be passed through the generating electrode system (electrodes 1 cm apart, in 10^{-1}M KNO_3). The current passed by the generator decreased as the electrolyte concentration was reduced following the increase in solution resistance.

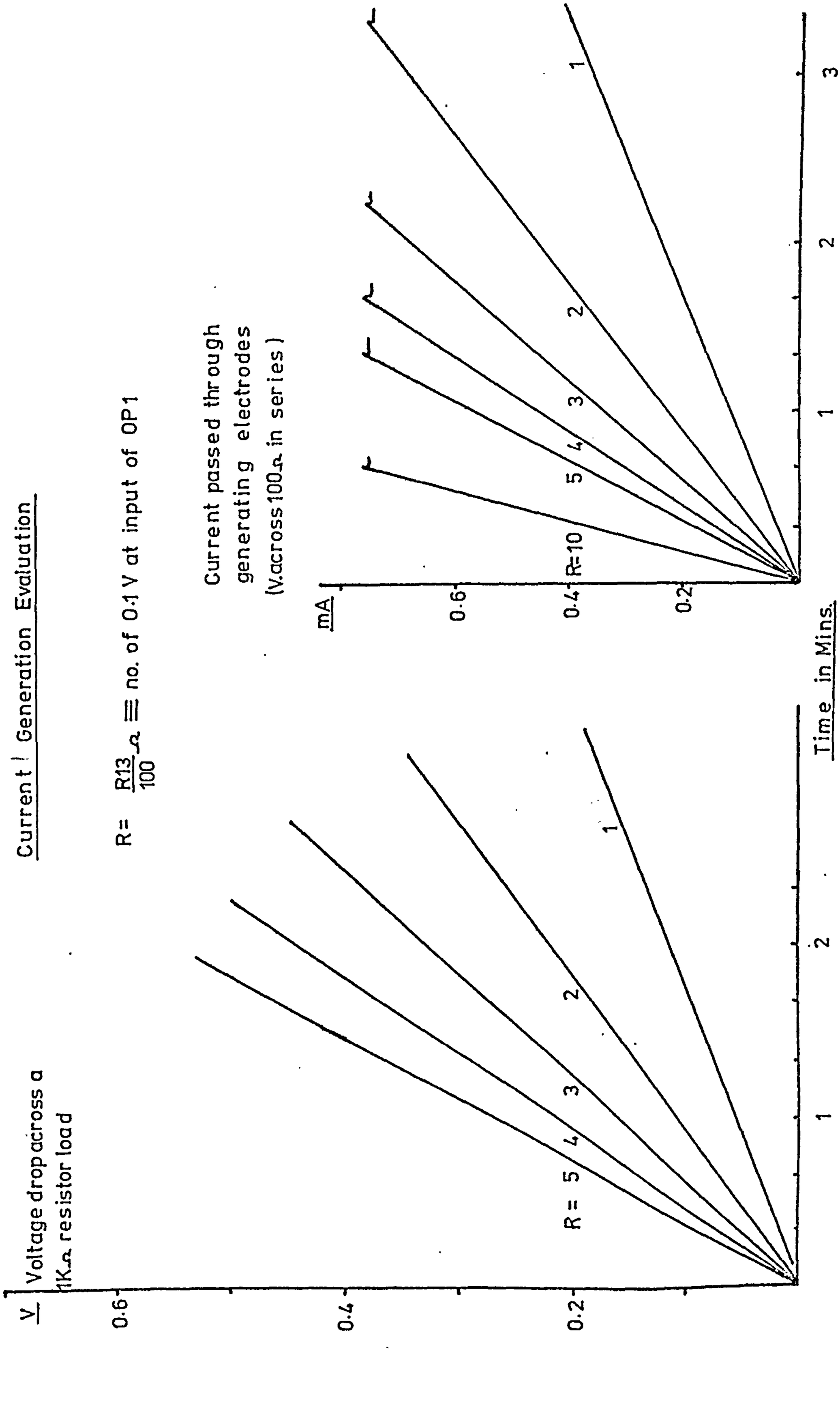
In the evaluation of silver generation efficiency, the membrane potential of a silver sulphide electrode was recorded as the ramp was operated at various gradients, (Figure 20.2.-II). The relationship between membrane potential (E) and silver activity (a) (and thus current (i)) is logarithmic and so in order to evaluate the linearity of silver generation an arbitrary scale was used on the potential axis and values of $\log^{-1}E$ *vs.* t($\propto$i) were plotted in Figure 20.2.-III, where t, time from initiation of gradient, is a measure of the current flowing (i). These plots are linear and show that the current generator can provide a linear concentration gradient of silver ions in solution.

20.2.2. Application of the Coulometric Gradient Generator to the Titration of Chloride Ions

Initial work,carried out on titration analysis using coulometric generation of silver,showed that in neutral and basic solutions silver ions were either not

Figure 20.2.-1

Current / Generation Evaluation



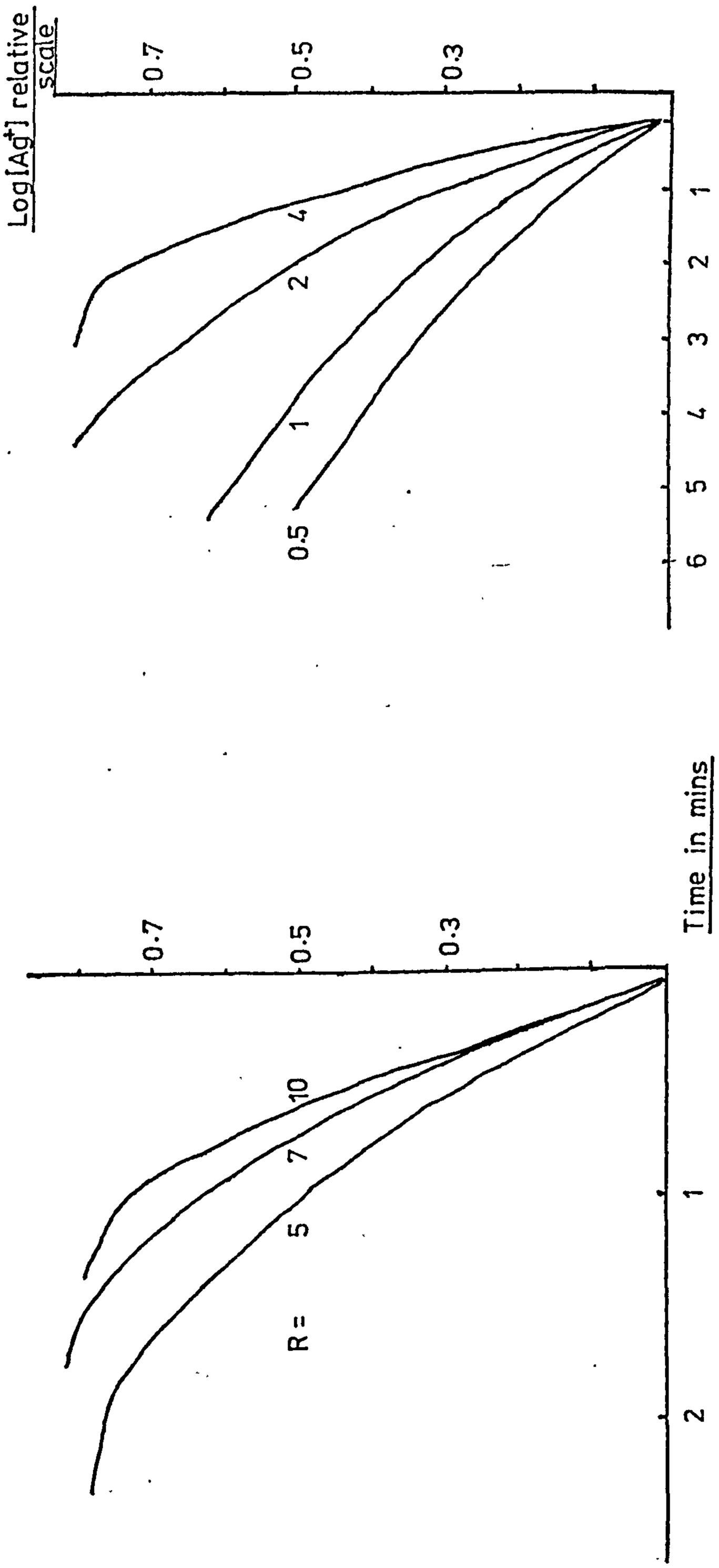
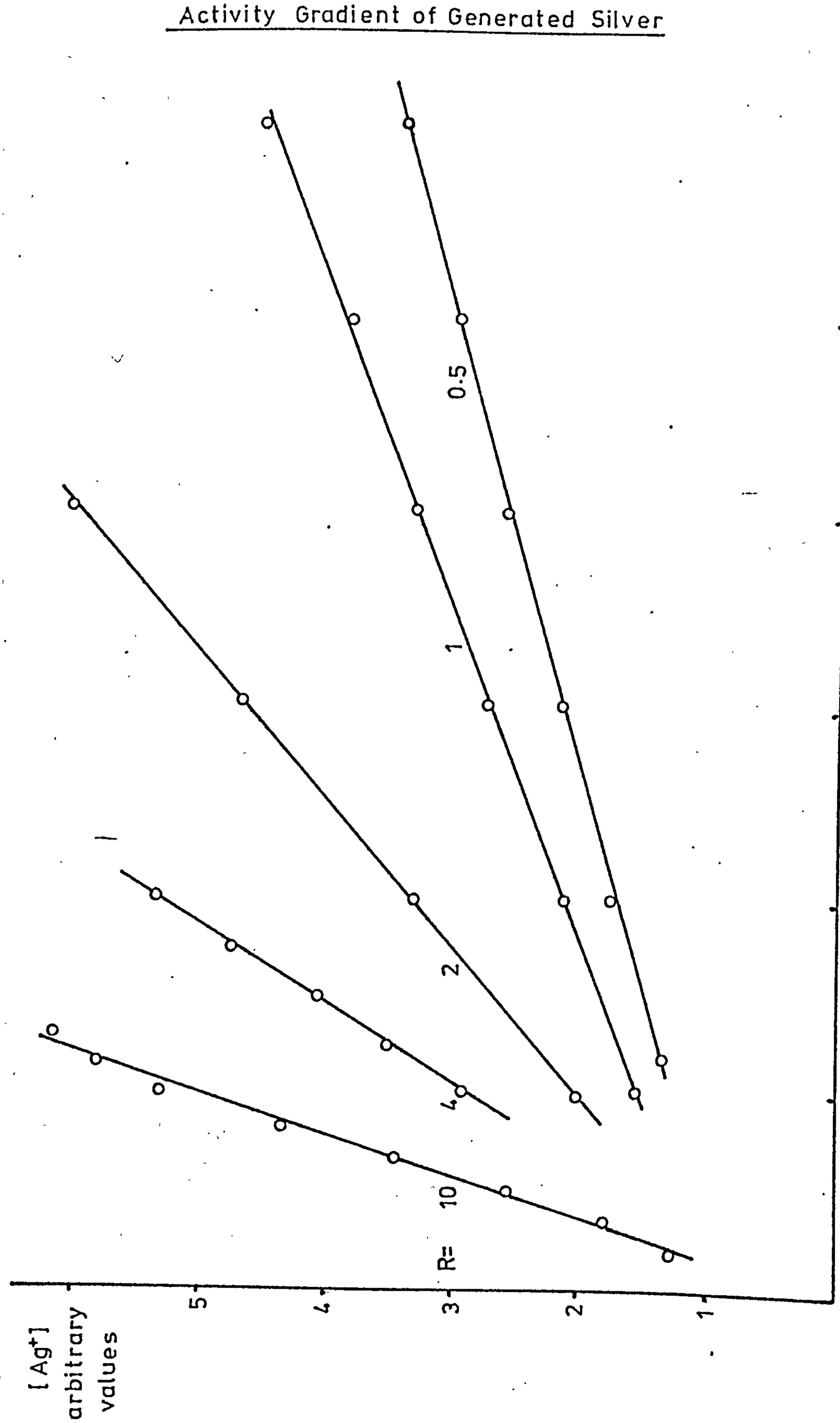


Figure 20. 2.-II

Electrode Response to Coulometrically Generated Silver

Figure 20. 2.-III

Activity Gradient of Generated Silver



released from the generating anode or rapidly formed silver oxide. In both cases the electrode was rendered passive after a few seconds. Dilute nitric acid effectively prevented the formation of coatings on the electrode surface but was sufficiently active to release silver ions by dissolution. These factors led to the successful adoption of the procedure suggested by Linnet (161), where 0.4 M sulphuric acid is used as a background electrolyte.

Potential-time curves for the titration of chloride solutions to a sulphide membrane /S.C.E. indicating system are given in Figure 20.2.-IV. The endpoint, taken as the maximum point of inflection, is distinct but difficult to locate visually without any electronic processing. Titration of the same solutions using the differential chloride/sulphide membrane detection system gave a vastly improved response (Figure 20.2.-V).

The values of elapsed time for samples in the range 0.05 to 0.3 mM, which include the dead time, are shown in the calibration graph, Figure 20.2.-VI. A value of the chloride concentration can be obtained from the measured titration time (elapsed time) and the calibration graph, or directly if the current efficiency and dead time of the system are known. It has already been shown that the current flowing at the equivalence point of a titration can be expressed as:

$$\frac{n.f.M.z.F}{6x104} \quad \text{amps}$$

where n = no of electrons involved in electrogeneration, f = flow rate in mls/min, M = concentration of sample in moles/litre, z = stoichiometric ratio of titrant to

Figure 20. 2.-IV

Coulometric Gradient Titrations of Chloride to a Sulphide Electrode Endpoint

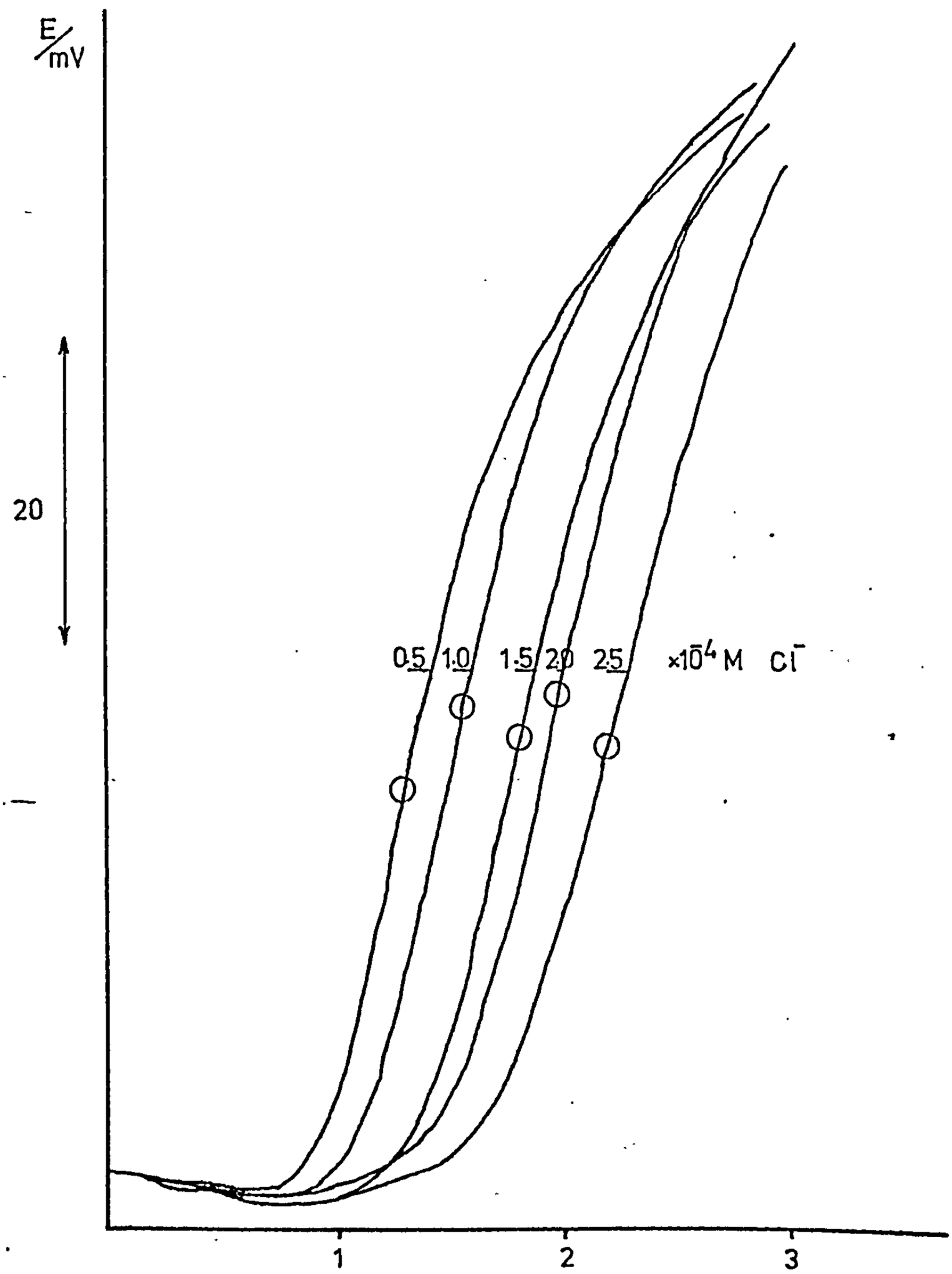


Figure 20. 2.-V

Coulometric Gradient Titrations of Chloride to a Differential Electrode Endpoint

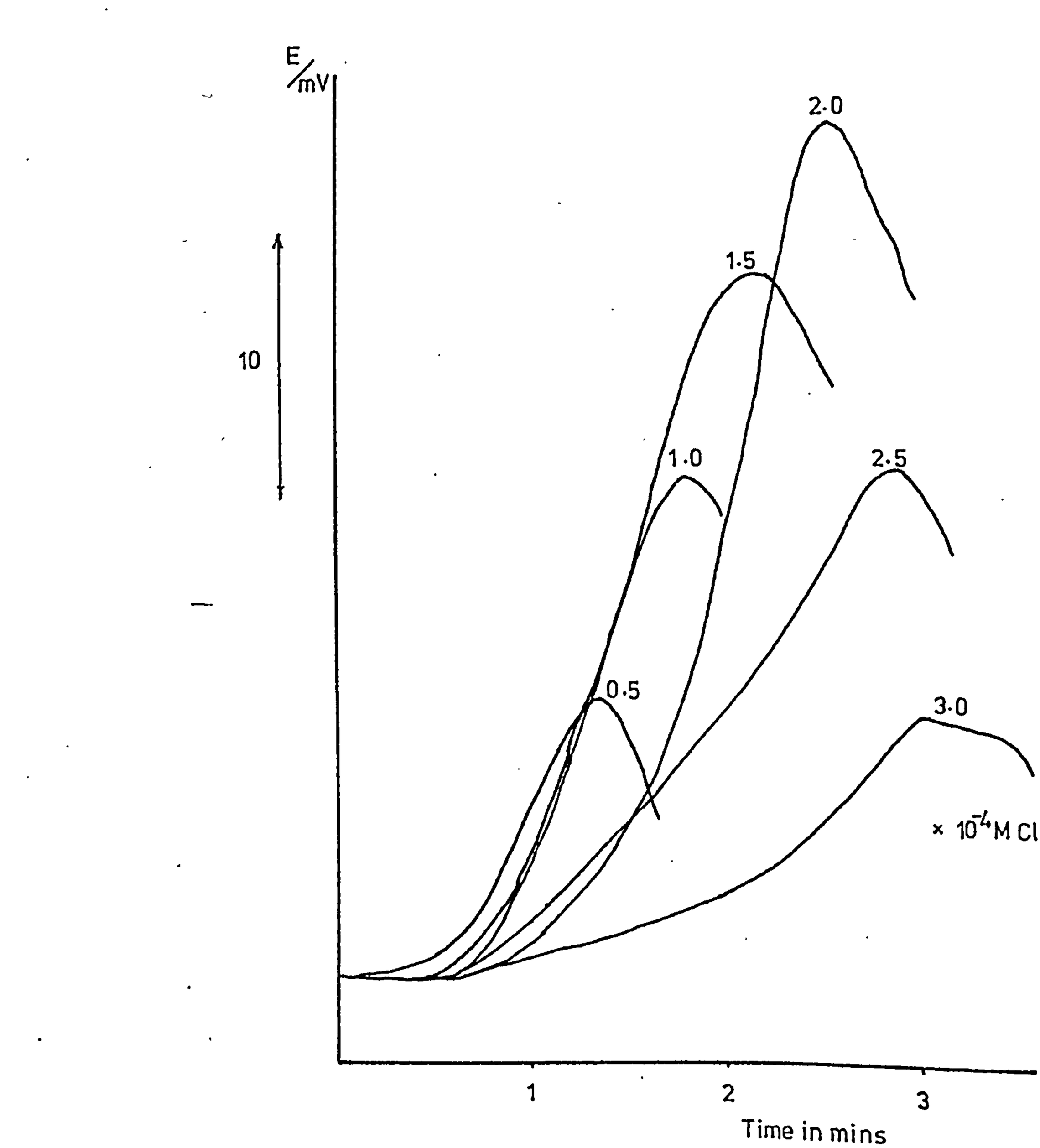
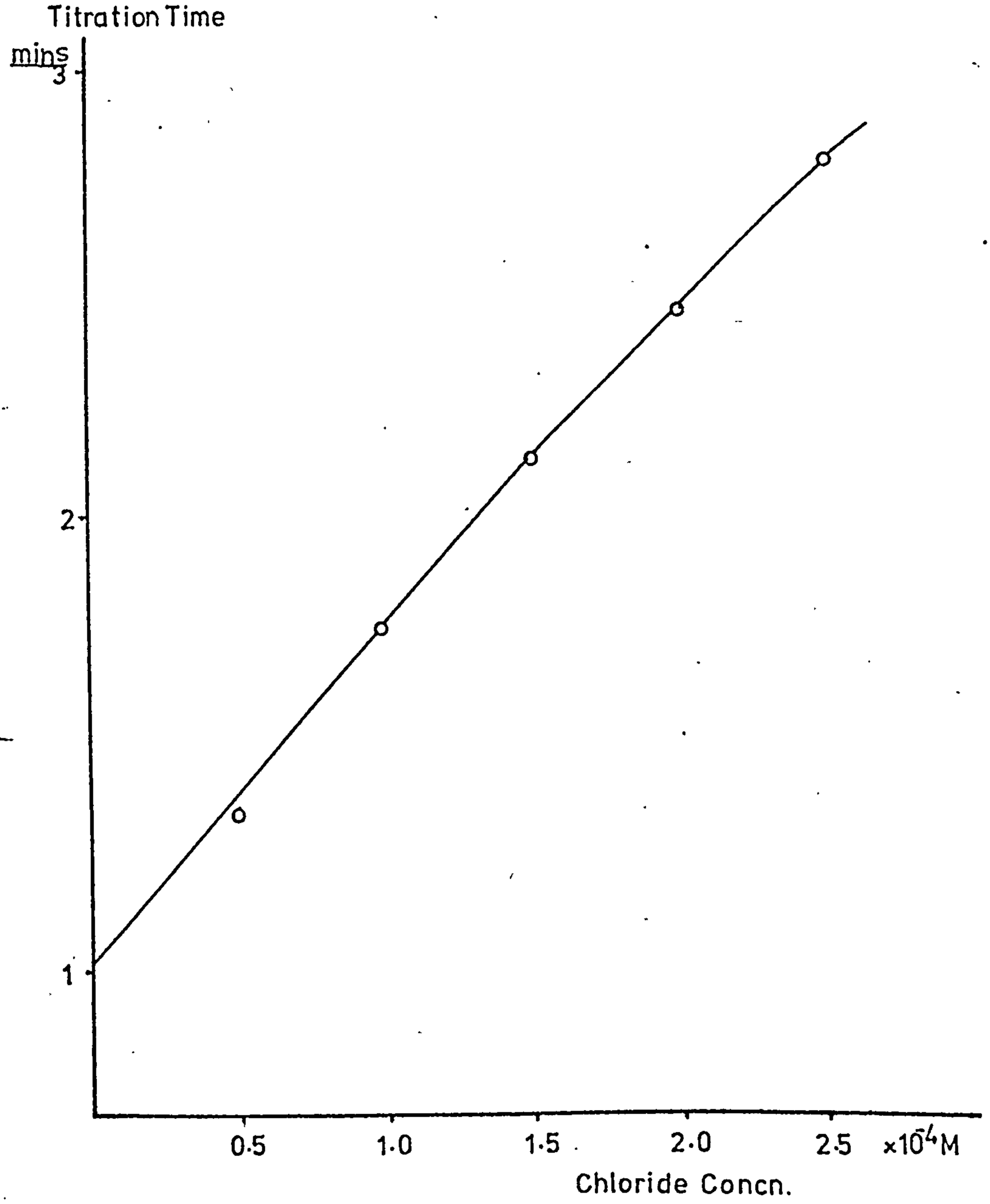


Figure 20. 2.-VI

Calibration of Coulometric Gradient Titrations



sample and F is the Faraday constant.

In the case of this titration for the 1.0×10^{-4} molar standard chloride solution : n=1, f=0.675, z=1 and assuming 100% generating efficiency;

Time to reach endpoint = 1.78 mins

Dead time (from calibration graph) = 1.06 mins

Titration Time = 0.72 mins

Current at endpoint = 108.0 μ A

Conc of std by calculation = $0.99(5) \times 10^{-4}$ M

Std. Concn. = 1.0×10^{-4} M

Calculated Concn. = $0.99(5) \times 10^{-4}$ M

20.3. Conclusions

The advantages of the gradient titration method for routine ion selective electrode potentiometry have already been outlined. The technique, in particular the coulometric generation mode also shows considerable promise for use in industrial process control.

In quality control applications the enhanced precision of gradient titration compared with direct potentiometry is of great importance. Furthermore the range of measuring titrations is increased since the sensing electrode may be responsive to the sample, reagent or reaction products. Many industrial monitors call for a real time measuring system which delivers a signal proportional to the concentration of the sought ion, coupled with some form of alarm system set to certain permissible levels. By utilizing short titration times, the gradient technique can be used to simulate a real time sensor while

the elapsed time interval can be used to activate a limit alarm.

The titration system could be fully automated by incorporation of a peak follower in the differential indicating mode. The follower, or null point crossing detector of a further differentiated signal could be utilized to sense the endpoint, stop and reset the current ramp and an integrator,to give a digital display of the titration time prior to activating the sampler. In this form the coulometric generated gradient titrator would form a useful fully automated analytical tool, for which the ion selective electrode is an ideal sensor.

CHAPTER 21

Automatic Analysis Of Divalent Metals, Using The Gradient Titration Technique

21.1. Titrations of Lead to a Virginiamycin Electrode Endpoint

21.1.1. Experimental

21.1.2. Results and Discussion

21.1.2.1. Precipitation Titrations

21.1.2.2. Complexation Titrations

21.2. Titrations of Calcium and Water Hardness with E.D.T.A. to an Ion Selective Electrode Endpoint

21.2.1. Experimental

21.2.2. Results and Discussion

21.2.3. Conclusions

21.1. Titrations of Lead, to a Virginiamycin Electrode Endpoint

The virginiamycin electrode was evaluated as an endpoint sensor for the single cup gradient titration of lead with both precipitating and complexing reagents.

As copper is a serious interferent for the Orion electrode and cannot be used as a titrant with this endpoint monitor, a solution of copper sulphate was used to titrate lead nitrate to the virginiamycin potentiometric endpoint. In the case of complexation titrations of lead, both E.D.T.A. and C.D.T.A. were used as titration reagents.

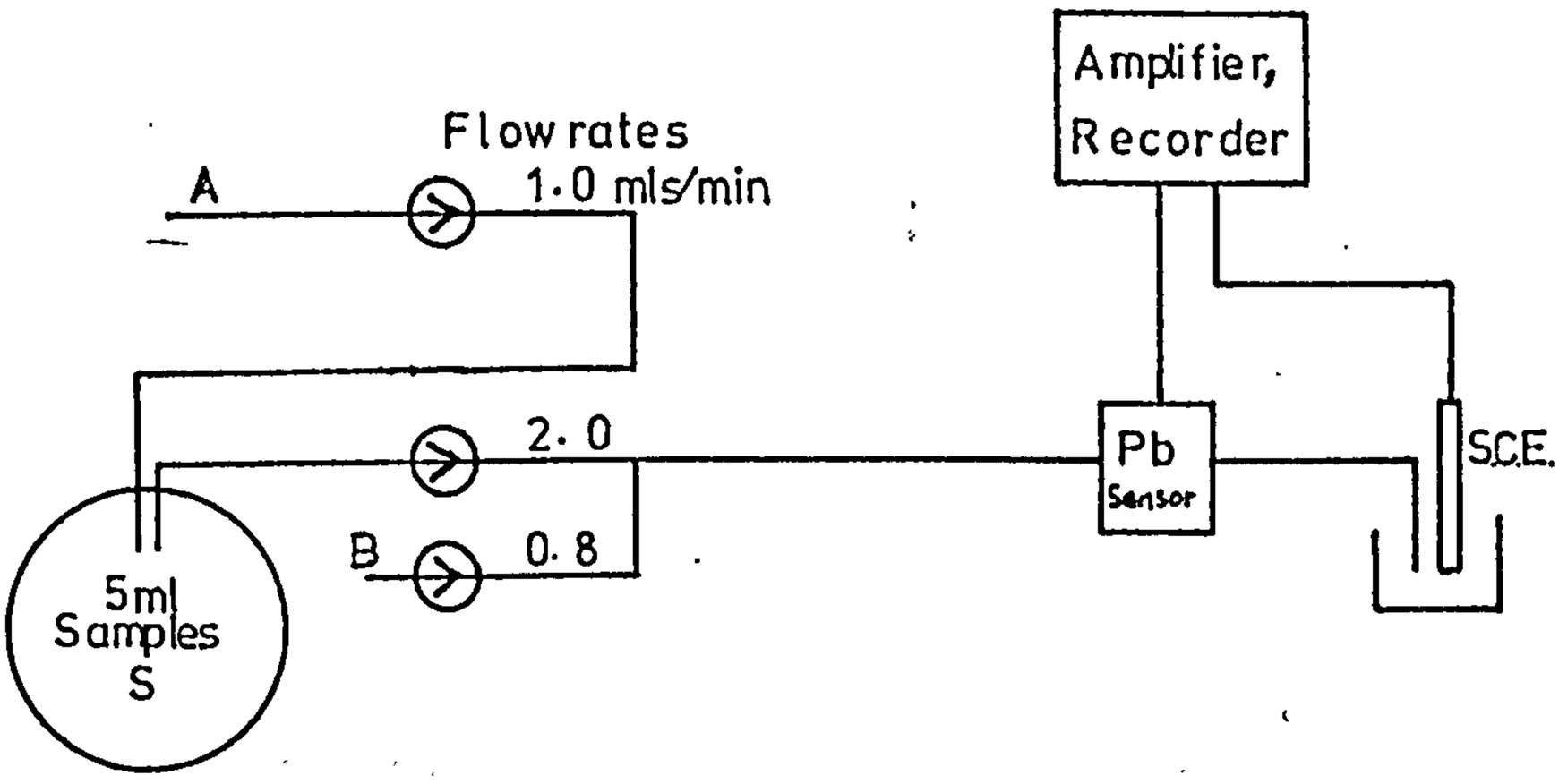
21.1.1. Experimental

The manifold design for the single cup gradient titration technique is shown in Figure 21.1.-I. In the

titrations of sulphate, lead nitrate (10^{-1}Molar) was used as a titrant and copper sulphate solutions were presented to the titration system by means of a Technicon sample turntable. As the ionic strengths of both solutions were high and no pH requirements had to be met, no buffer solution was used. In the case of the titrations of lead with the complexing agent, lead nitrate samples were placed in the turntable, and a buffer stream of ammonia, ammonium chloride (pH 10), with tartaric acid to prevent precipitation of lead hydroxide, was mixed with the post-titration stream before it reached the measuring cell.

Figure 21.1.-I

Manifold Design for the Titration Analysis of Lead



Sulphate Titration

A, $Pb(NO_3)_2$ 10^{-1}M

S, $CuSO_4$

B, ——

Complexation Titration

A, E.D.T.A./C.D.T.A.

S, $Pb(NO_3)_2$

B, 0.5M NH_3/NH_4Cl
+ Tartaric Acid

21.1.2. Results and Discussion

21.1.2.1. Precipitation Reactions

A typical potential/time curve for the titration of sulphate is given in Figure 21.1.-II.

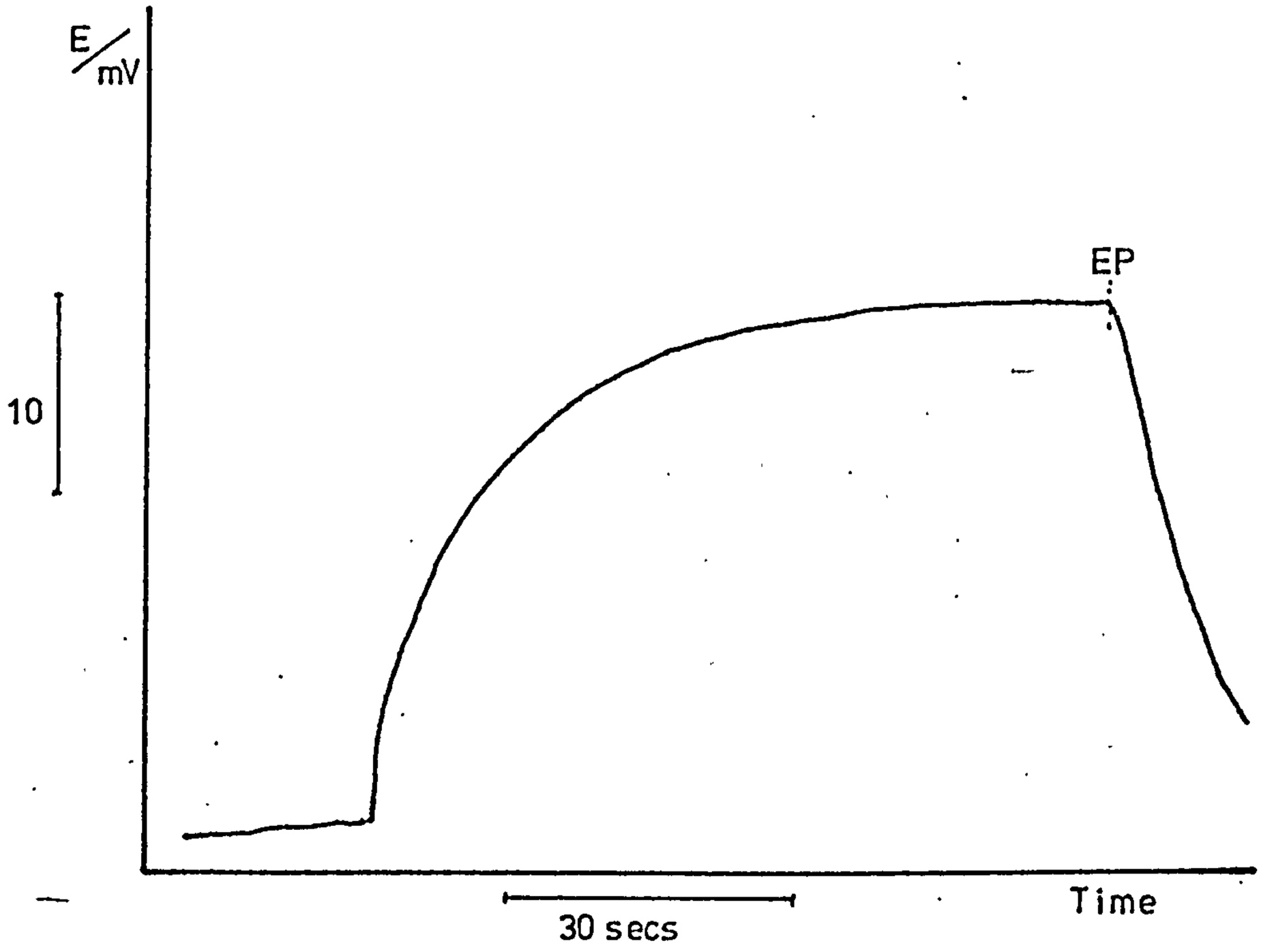


Figure 21.1.-II

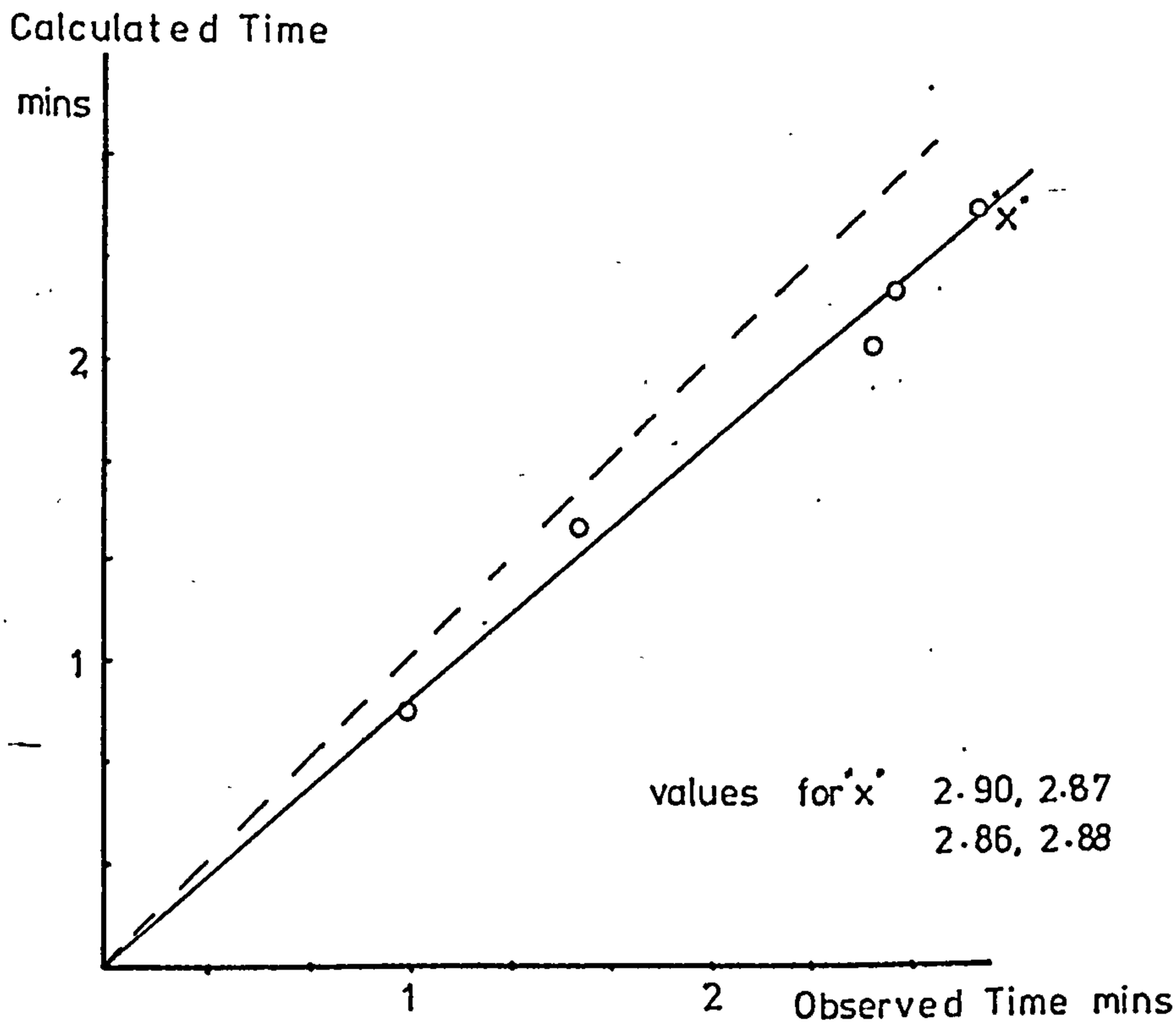
Titration Curve for 4×10^{-3}M SO_4^{2-} vs 10^{-1}M Pb^{2+}

The break (EP) in the curve was initially assumed to be the endpoint of the titration. The validity of this assumption was investigated by titrating several different concentrations of sulphate and plotting the 'observed' end-point against the calculated endpoint. The reproducibility of the titration was evaluated by repeating the analysis of several samples of the same solution. It can be seen from

these results (Figure 21.1.-III) that the observed end-point occurs after the calculated equivalence point, but the titration is reproducible and can be carried out, in conjunction with a calibration graph, by an automatic system using the lead selective electrode as an endpoint sensor.

Figure 21.1.-III

Titration Endpoint Evaluation



The major drawback with this system, especially at this high concentration, is the formation of a heavy precipitate as the titration product. The precipitate can easily disrupt the flow in the pump tubing to give a large error in the analysis and may cause complete blockage resulting in failure of the titration system. Although

filtration is possible in an automated system, the amount of mixing that would occur is unacceptable; however, a well designed flow cell with no 'sharp corners' or sudden changes in tube diameter should allow the precipitate to pass through the system without blocking.

When the concentrations of the solutions are low and the cell design is good, automated analysis of sulphate can be achieved using ion selective electrodes as endpoint sensors but the analysis of lead can be improved by titration with a complexing agent.

21.1.2.2. Complexation Titrations

A potential vs. time plot for the titration of lead with E.D.T.A. is given in Figure 21.1.-IV. Although in a non automated method of analysis an endpoint is clearly defined, there is no such point in the results for the gradient titration and thus the virginiamycin electrode cannot be used as a sensor in the gradient titration of lead with E.D.T.A.

The experiment was repeated with a C.D.T.A. titrant and the titration curve (Figure 21.1.-V) shows a potential break that can be related to an equivalence point. The validity of the break as an endpoint was tested by repeating an individual titration to check reproducibility and titrating a series of lead concentrations to evaluate the relationship between the observed endpoint and the calculated equivalence point. The results (Figure 21.1.-VI) indicate that although the observed potential break occurs after the equivalence point, it can be used for analytical purposes provided that the sample concentration is evaluated from a

Figure 21. 1.–IV

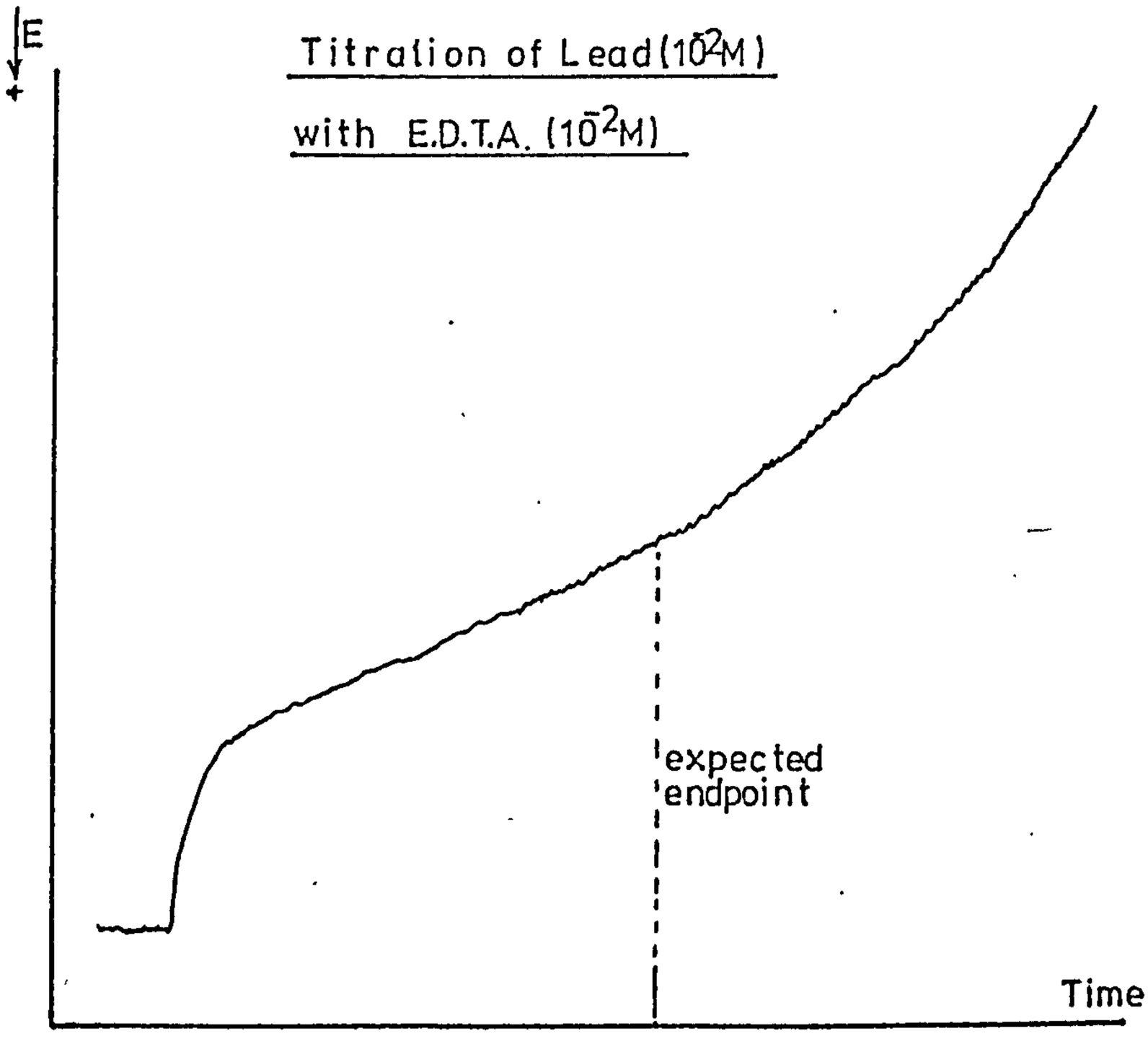
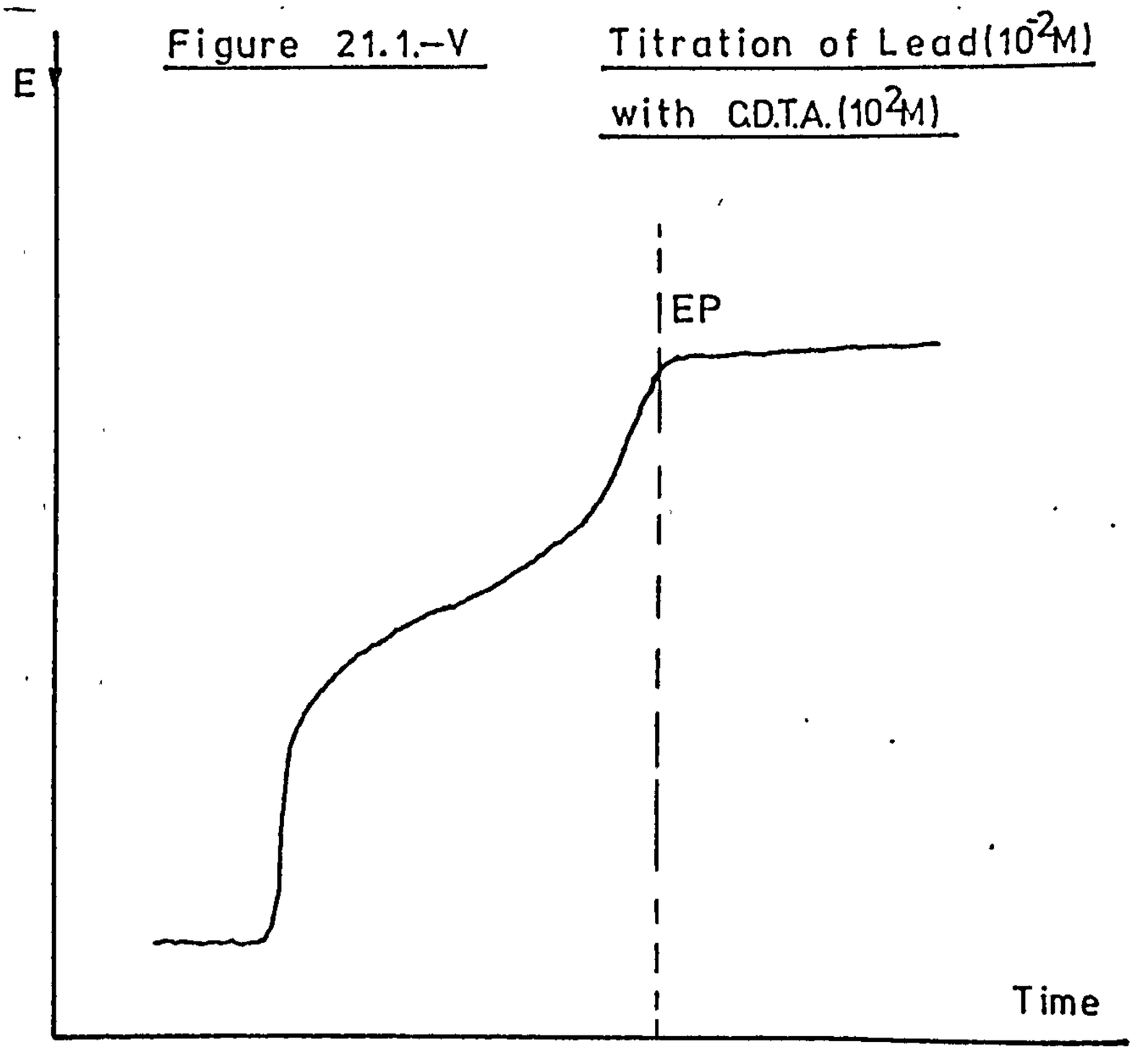


Figure 21.1.–V



calibration graph . The deviation of the calibration graph from the theoretical value is due to a slight non-linearity of the concentration gradient and it was found that Technicon pump tubes could not be calibrated with sufficient accuracy to ensure a perfectly linear gradient over a period of ten minutes. In addition to this, the slow response time of the electrode is evident at the beginning of the titration and probably causes some delay in the endpoint sensing.

21.2. Titrations of Calcium and Water Hardness with E.D.T.A. to an Ion Selective Electrode Endpoint

The calcium ion selective electrode has frequently been used for automatic titrations of both calcium (162) and calcium plus magnesium (163) in the measurement of water hardness. The endpoint detection in both of these titrations relies on the change in potential of the electrode as calcium is removed from solution.

When the analysis of total divalent metal in solution is required, especially in the absence of calcium, an electrode responsive to all divalent metals must be used as the endpoint sensor. In this work the water hardness electrode was compared with the calcium electrode, and used as an endpoint sensor in the automated titration of water hardness.

It has been shown that the most significant deviation of the titration calibration graphs from their theoretical shape is caused by the slight non-linearity of the concentration gradient. In the case of titrations

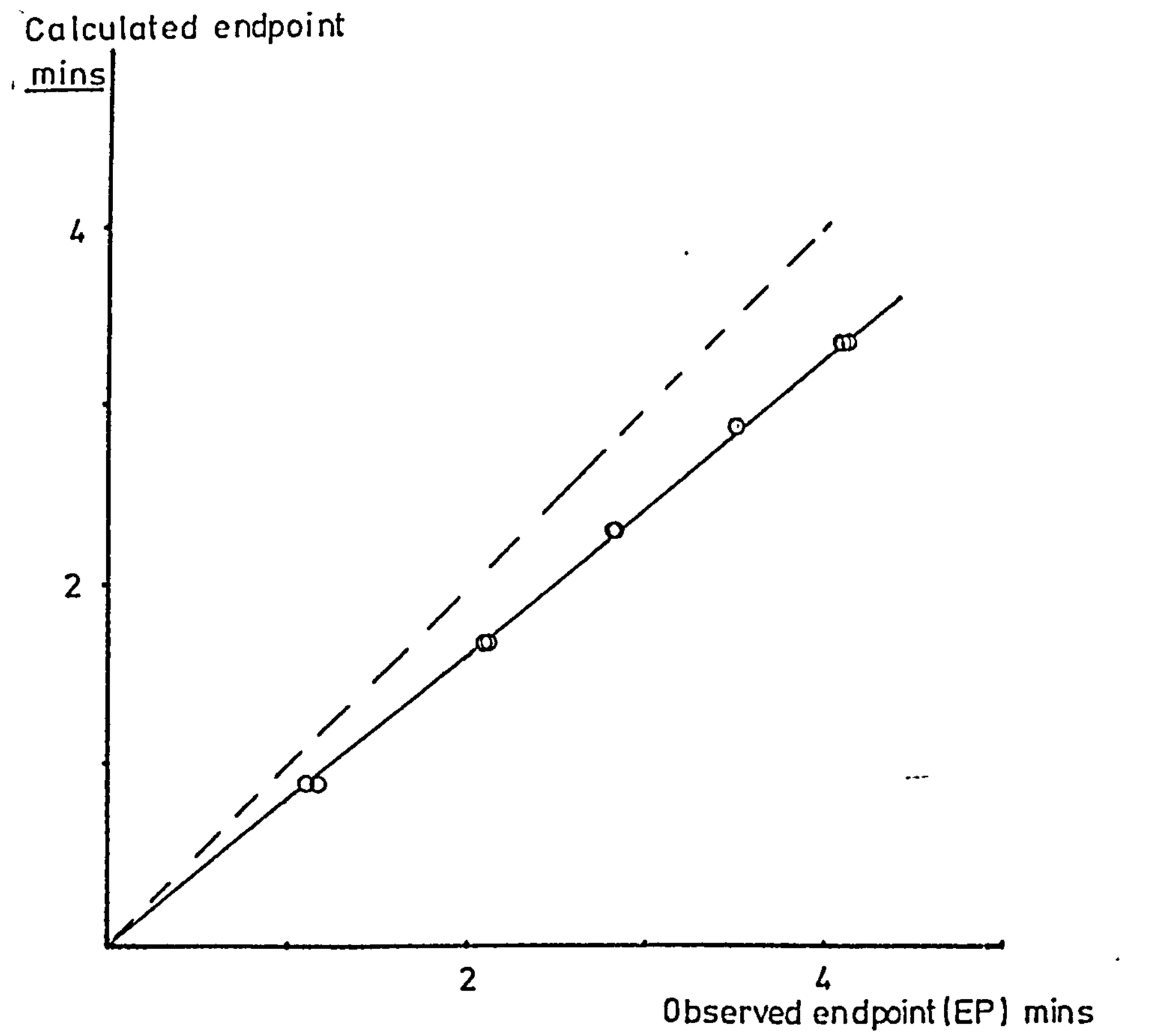
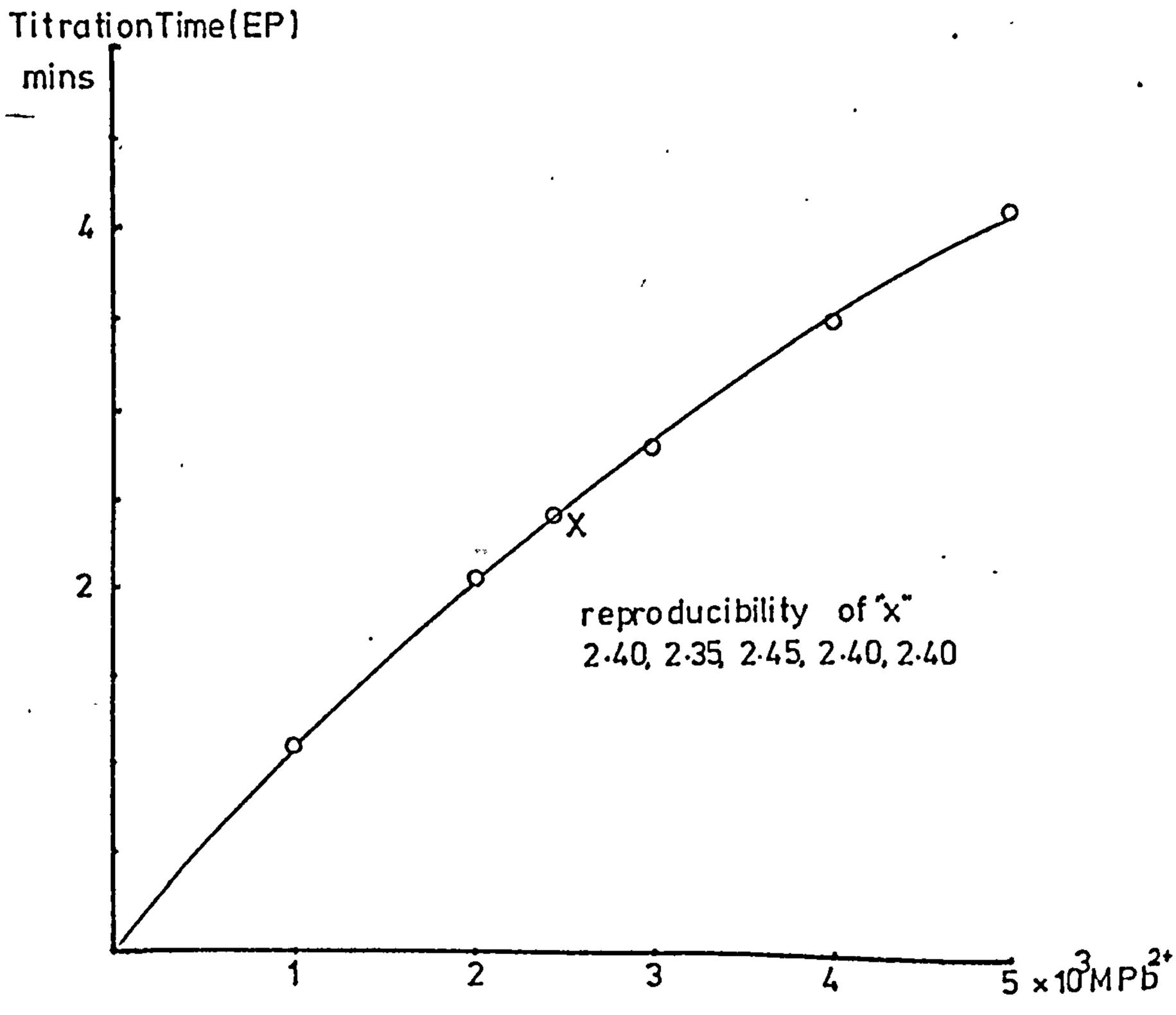


Figure 21.1.–VI Lead (10mls) vs CDTA(10^{-2}M) Titration Endpoint Analysis



to a liquid membrane electrode the 'rounded' endpoints also cause some difficulty in positioning the equivalence point with a high degree of reproducibility. In an attempt to reduce these effects to a minimum, a piston pump was designed to give an accurately reproducible gradient, and electronic differentiators were made to provide a doubly differentiated signal to aid the analytical measurements.

21.2.1. Experimental

A piston pump was designed and constructed to generate a linear gradient for the single cup technique, in order to avoid use of the expensive Technicon Auto-Analyzer equipment and variations in flow rates due to ageing of the tubes. The pump, shown in Figure 21.2.-I, has two chambers, A, B, one of which (A) has a volume twice that of the other. When the piston (p) is moved with constant speed along the chamber,the flow rates of solutions from A and B will be in the ratio 2:1, and of a magnitude proportional to the velocity of the piston. The pump was evaluated by measuring the net decrease in weight of a sample container as solution was abstracted at 2F mls/min and added at F mls/min. A linear decrease in weight with time would indicate a constant flow rate.

Two operational amplifier based differentiators were made to process the electrode signal. A circuit diagram for the apparatus is shown in Figure 21.2.-II. This allows both the first and second derivatives of the potential vs. time graph ($\frac{\partial E}{\partial t}/t, \frac{\partial^2 E}{\partial t^2}/t$) to be recorded with the original signal (E vs. t) on a three channel chart recorder.

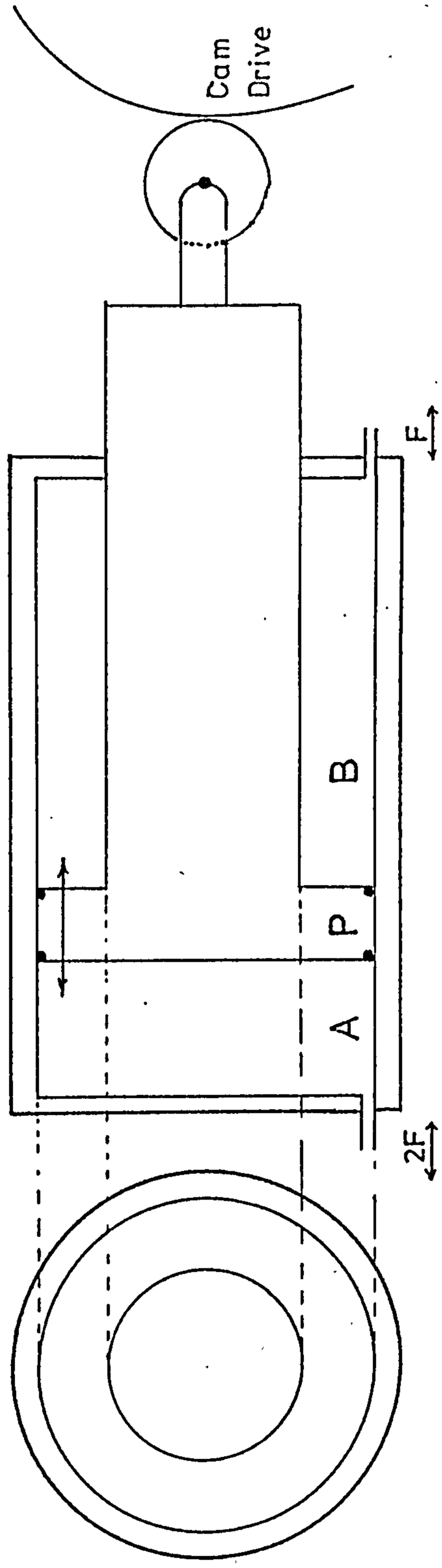
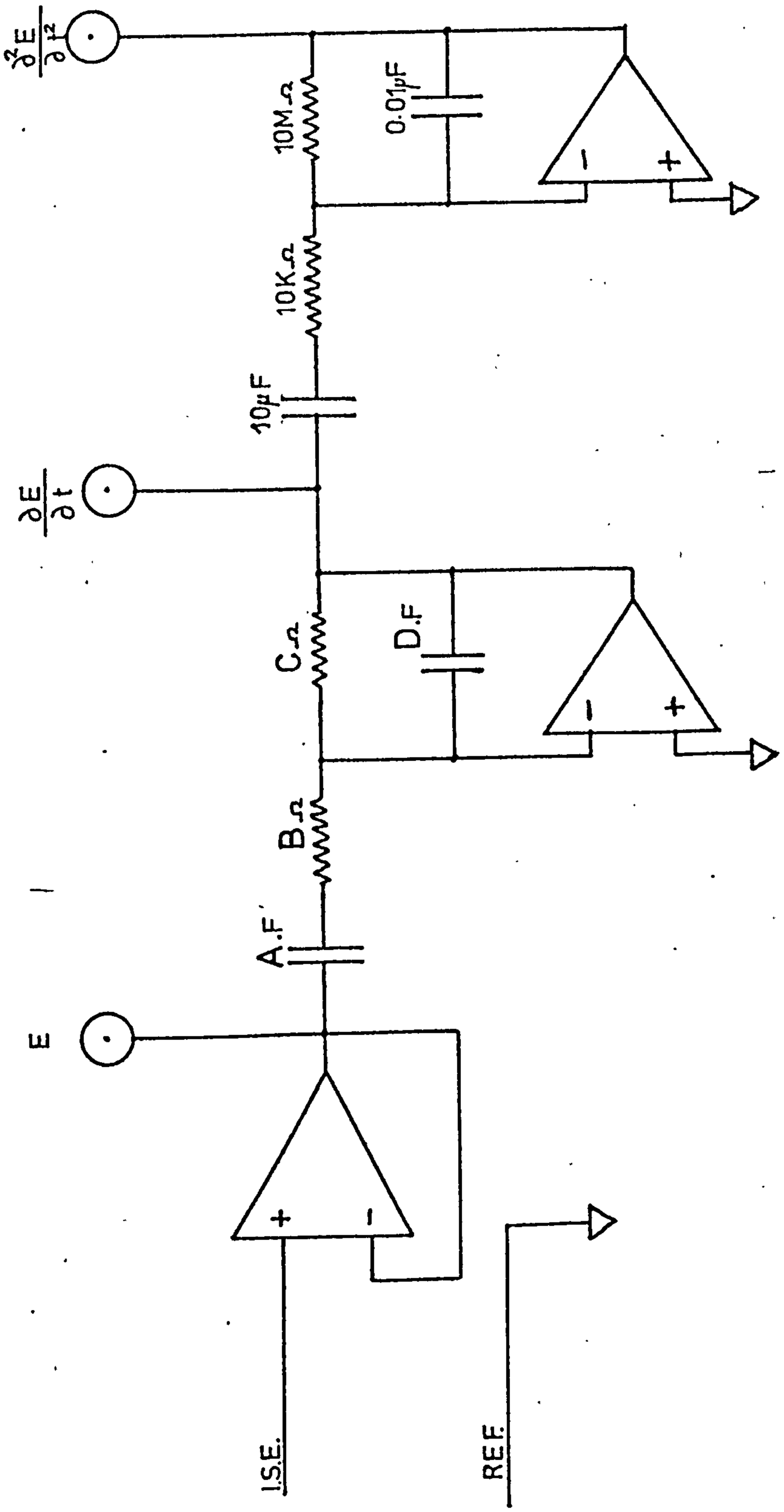


Figure 21.2.-I Piston Pump Design

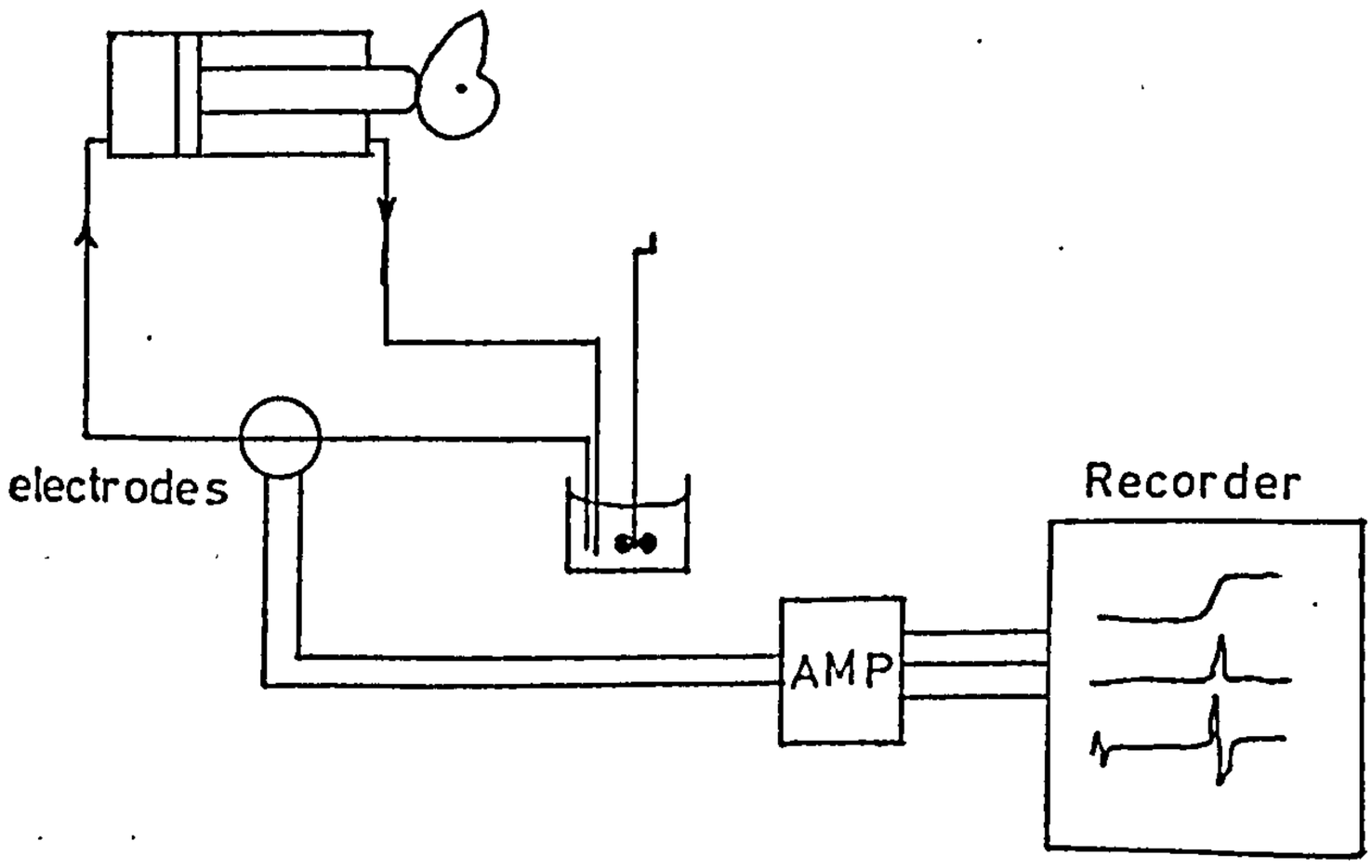
Figure 21. 2.-II

Electrode Signal Differentiator



Samples of calcium solution buffered to pH 10 with an ammonia, ammonium chloride solution were titrated to a calcium electrode potentiometric endpoint with E.D.T.A. The recorder trace of potential vs. time was analysed and an endpoint was obtained and compared with the theoretical value using the experimental arrangement shown in Figure 21.2.-III. Titrations of the calcium solutions were repeated while all three electronic functions were recorded (E vs. t, $\partial E/\partial t$ vs t, $\partial^2 E/\partial t^2$ vs t) and the results were analysed for both calcium and water hardness electrodes. A series of solutions containing barium, magnesium, calcium, and mixtures of these ions were prepared and analysed to investigate the use of the water hardness electrode as a sensor for the automated titration of divalent metals in solution.

Figure 21. 2.-III



Titration Manifold Design

21.2.2. Results and Discussion

Evaluation of the Pump

The volume of solution expelled from, or drawn into each half of the pump during a titration cycle was measured. The volume of A was 20 mls, and the volume of B was 10 mls. The titration time was 96 seconds with a reset time of 24 seconds. Thus the complete cycle takes two minutes. A plot of the mass change of sample and container against time,during a titration period, (the results for which are given in Table 21.2.-IV) is linear, indicating that the flow rates are constant.

Table 21.2.-IV Mass Change of Sample and Container during a Titration Cycle

Time in Seconds	0	10	20	30	40	50
mass in grams	25.19	24.14	23.15	21.98	20.84	19.58
seconds	60	70	80			
grams	18.70	17.66	16.70			

It can be concluded from these results that the two flow rates are constant and in the ratio 2:1.

Titration Analysis

A typical potential curve for the titration of calcium is shown in Figure 21.2.-V. Initially the break (E.P), was assumed to be the electrode endpoint and the validity of this assumption was evaluated by plotting a graph of assumed endpoint vs. calculated equivalence point for a series of calcium solutions (Figure 21.2.-VI). The good correlation between the observed and calculated endpoints indicates that this is a viable analytical technique but as

the concentration of calcium is decreased, and as the selective membrane ages, the endpoint becomes less well defined. After three days use as a titration endpoint indicator, the response of the membrane had been degraded to the curve shown in Figure 21.2.-VII, with its first and second derivatives. Although it is possible to extract an endpoint from this curve (E vs. t), it is unlikely that any accuracy could be achieved. In the case of the second derivative, however, the zero crossing provides an easily identifiable position on the recorder chart to which the endpoint of the titration can be related. The results for the titrations of calcium solutions, measuring between the zero crossings, are given in Figure 21.2.-VIII for both the calcium and water hardness electrode endpoints. The elapsed times for the individual electrodes agree with each other very well, but they lag behind the theoretical values. This effect, however, was also observed by Hadjiioannou (163) and does not cause any error, provided that the unknown concentration values are read from a calibration graph. The composition of the divalent metal solutions analysed, and their titration values, are given in Table 21.2.-IX.

Table 21.2.-IX Analysis of Divalent Metal Solutions

Sample No	Solution Taken (mls 10^{-1}M in 50 mls)					
	Ca	Mg	Ba	Total M^{2+}	Titn Time	M^{2+} Found
1	1.0	4.0	-	5.0	12.85	4.95
2	2.5	1.5	-	4.0	11.30	4.02
3	-	-	4.5	4.5	11.95	4.40
4	-	5.0	-	5.0	12.77	4.90
5	2.0	2.5	-	4.5	12.06	4.45
6	2.5	-	2.5	5.0	13.00	5.02
7	1.0	3.0	1.0	5.0	12.80	4.91

Figure 21. 2.-V

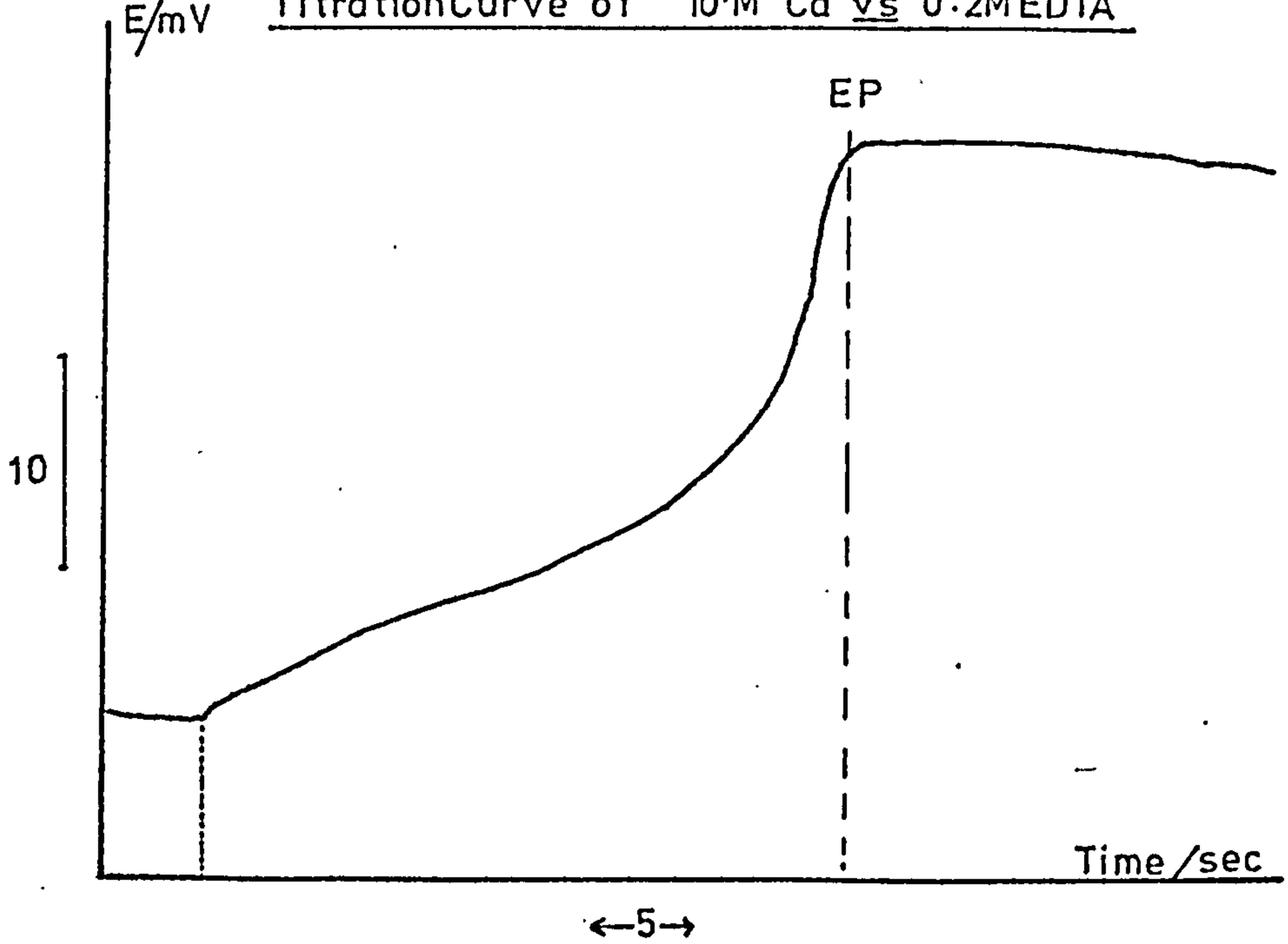


Figure 21. 2.-VI

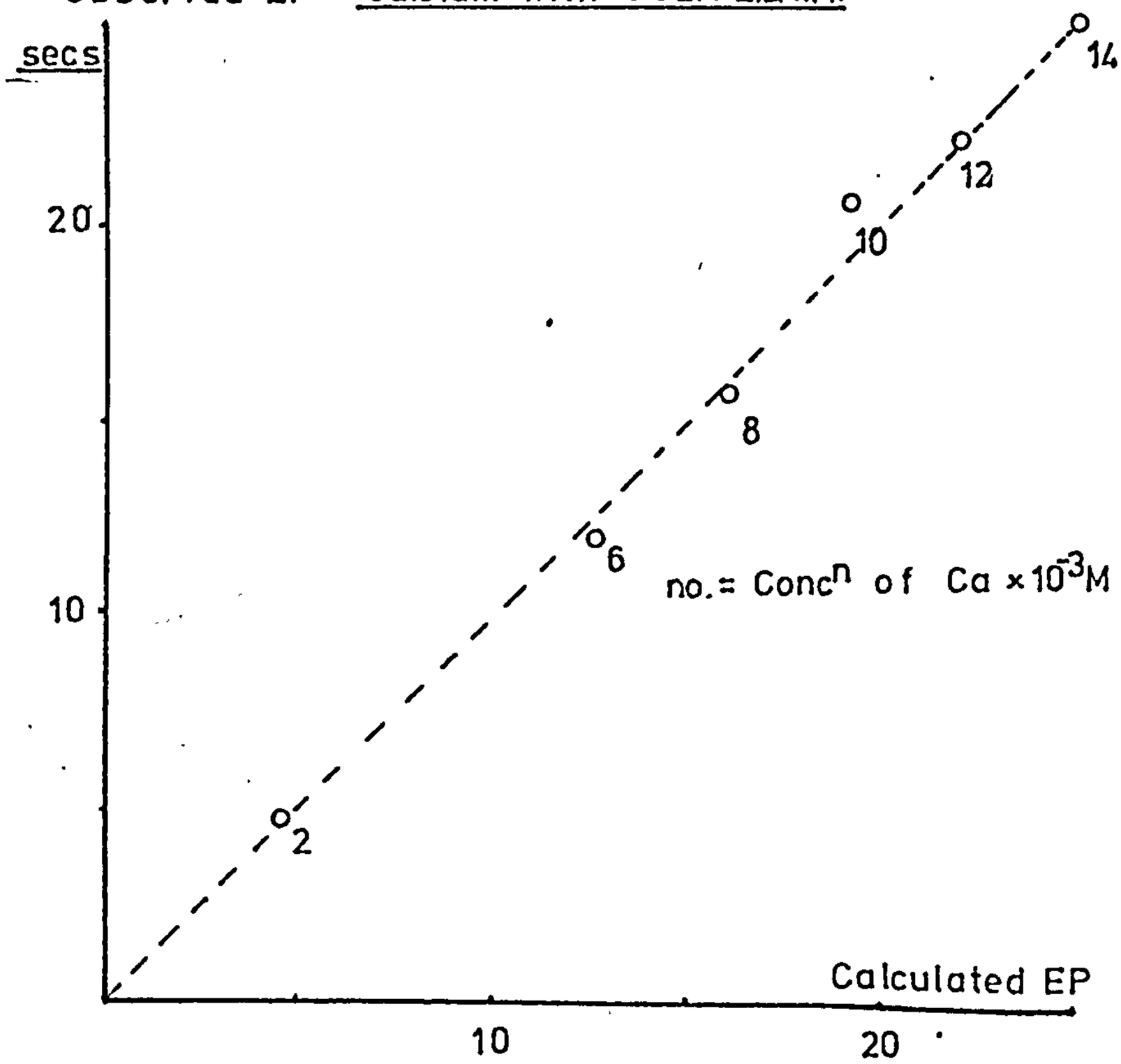


Figure 21.2.-VII

Recorder Trace of a Calcium(10^{-2}M) vs. E.D.T.A.(0.02M) Titration

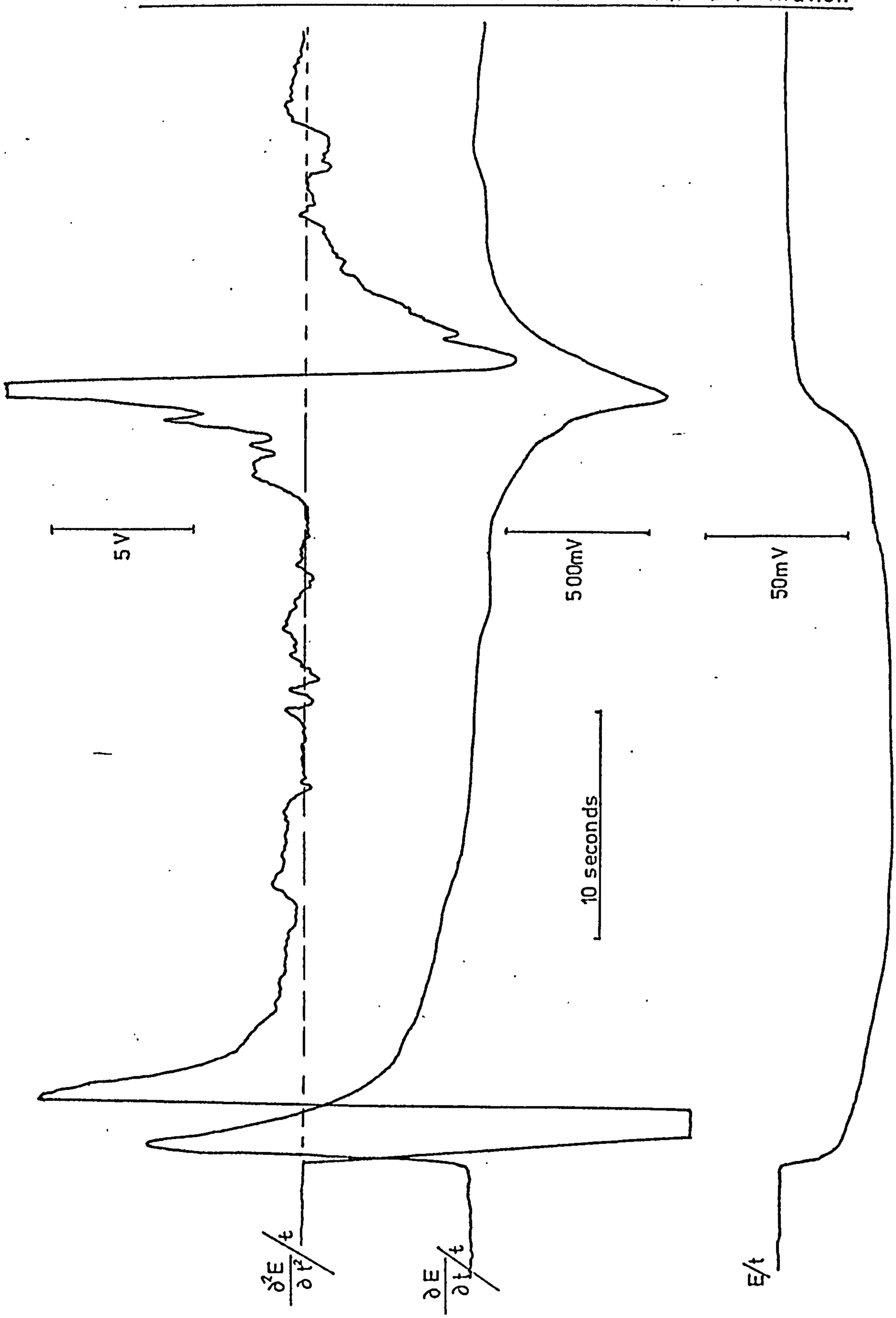
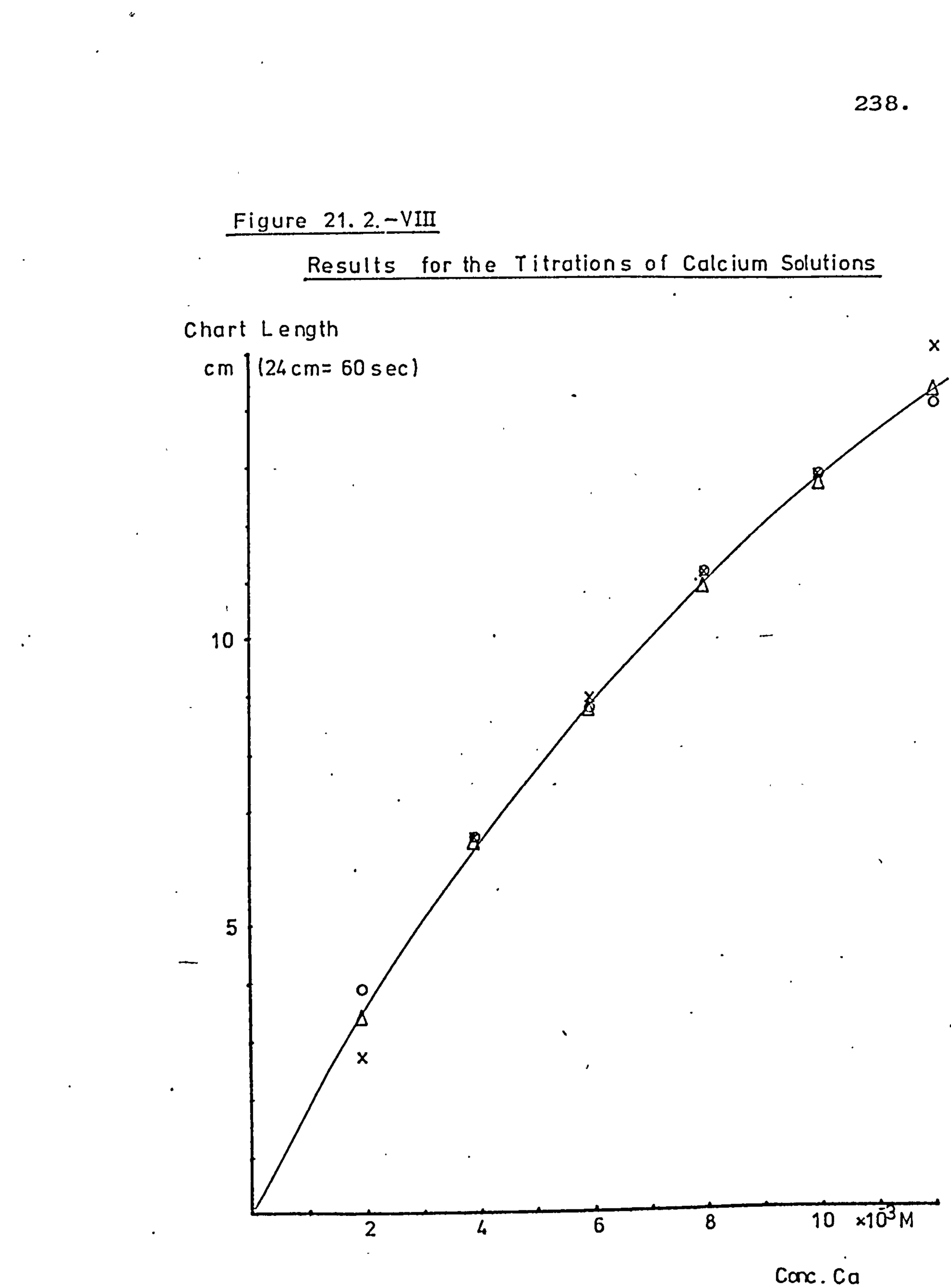


Figure 21. 2.-VIII

Results for the Titrations of Calcium Solutions



21.2.3. Conclusions

From these results it can be seen that an automated titration method of analysis can be carried out using the water hardness electrode as a potentiometric sensor. The main disadvantage of the method is that ammonium ions, used as a pH buffer, are an interferent for the electrode and cause a decreased potential jump at the endpoint. This can be avoided with some metals, e.g. calcium and magnesium, by using a glycine (pH 9.7) buffer, but in many cases the ammonia, ammonium chloride buffer is recommended for the E.D.T.A. titration (164).

The endpoint obtained with the doubly differentiated signal is more easily located than an inflection and the chart length between two zero crossing points provides a reproducible measurement of elapsed time during a titration. The zero crossing points also provide ideal electrical signals for the electronic measurement of titration time and the initiation of functions,such as sample changing,necessary for complete automation.

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APPENDIX

Computer program ADDFIT

The program was written by Brand and Rechnitz (88) to evaluate an unknown concentration (C) from the potential of an electrode (E_i) using the multiple standard additions technique (8.1.5).

Values of E_o, S and C are obtained by simultaneous solution of equation 8.1.-2 for a series of potential values obtained after additions of a standard solution (concentration C_i, volume V_i).

$$E_i = E_o + S \log \frac{CV + \sum_{i=1}^{j} C_i V_i}{V + \sum_{i=1}^{j} V_i} \qquad (8.1.-2)$$

after one addition;

$$C = \frac{C_2 V_2}{(V_2+V)\ \exp[2.303(E_2-E_1)/S] - V}$$

$$E_o = E_1 - S \log C$$

An error function, Z_i, is calculated from the equation,

$$Z_i = E_i = E_o - S\ x_i \quad , \quad (x_i = {}^{dE_i}/_{dS})$$

and values of ΔE_o, ΔS and ΔC are calculated by a least squares curve fitting procedure for the equation,

$$Z_i = \Delta E_o + \Delta S\ x_i + \Delta C\ y_i \quad , \quad (y_i = {}^{dE_i}/_{dC})$$

E_o, S and C are corrected by addition of the error values and the process is repeated until the values converge.

A complete listing of the program, in Fortran IV, is given in reference 88.