

**DEVELOPMENT OF AN OPTICAL
SOL-GEL-BASED
DISSOLVED OXYGEN SENSOR**

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DECLARATION

I hereby certify that this material, which I now submit for assessment on the programme of study leading to the award of Doctor of Philosophy, is entirely my own work and has not been taken from the work of others save and to the extent that such work has been cited and acknowledged within the text of my work.

Signed: Aisling McEvoy

Date: 23.09.1996

DEDICATION

To Mum and Dad

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ABSTRACT

A dissolved oxygen sensor based on the quenching of fluorescence from a ruthenium dye complex entrapped in a porous sol-gel film is reported. Sol-gel-derived silica films were fabricated by dip-coating onto planar and optical fibre substrates. The films were pre-doped with oxygen-sensitive ruthenium complex, $[\text{Ru(II)}\text{-tris(4, 7-diphenyl-1,10-phenanthroline)}]^{2+}$, whose fluorescence is quenched in the presence of oxygen.

The structure and behaviour of sol-gel films is dependent on the fabrication parameters. By varying these parameters, different microstructures and consequently different quenching responses to dissolved oxygen are obtained. In particular, enhancement of the surface hydrophobicity increases the quenching response in water. This is achieved by using suitable proportions of modified precursors of silica of the form $\text{R(OEt)}_3\text{Si}$, where R is an alkyl group, in the standard fabrication procedure. It is shown that by extending the aliphatic group of the modified precursors, the quenching response in aqueous phase increases. In this way, the film has been optimised for dissolved oxygen sensing.

The development of a dissolved oxygen sensor is mapped from the preliminary oxygen/dissolved oxygen measurements using a laser-based characterisation system through to a small, portable, all-solid state system, which can be used in different modes e.g. fluorescence intensity and phase fluorimetric modes, as well as two different configurations, planar- and fibre-based configurations, all of which have been demonstrated here.

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CHAPTER 1: INTRODUCTION

1.1 Introduction

A sensor is a device which measures the value of a physical parameter, or the concentration of an analyte. There are many different families of sensors such as mechanical, electrical, thermal, electrochemical etc., but the sensors of interest in this work are optical chemical sensors, in particular optical oxygen sensors. This body of work is based on the development of an optical sol-gel-based dissolved oxygen sensor. The porous sol-gel glass is doped with a ruthenium dye complex whose fluorescence is quenched by oxygen. The development of the sensor is traced from the preliminary materials investigations in gas phase, through the design and assembly of various characterisation systems, to the sensor optimisation for the development of a prototype instrument.

1.2 Dissolved oxygen and its importance

Water is a basic necessity of every living thing, animal, plant or organism, on the earth. The beneficial uses of water are varied and include its use for drinking and for domestic purposes, for watering livestock and the irrigation of crops, for fisheries, both game (salmonid) and coarse (cyprinid), for industry and for food production, for bathing and for recreational and amenity use. If water is rendered unsuitable for any of these purposes, then it is polluted to a greater or lesser degree depending on the extent of the damage caused. Pure water i.e. water which is absolutely free from any substances dissolved in it, does not exist outside the laboratory, as natural water contains dissolved gases and salts. These substances are often the very factors which make the water suitable for particular beneficial uses. Often dissolved oxygen (DO) is the constituent of water which is monitored to give an indication of the water quality.¹ The amount of DO in natural waters is not static. It is rather in a state of dynamic equilibrium with a delicate balance maintained by biochemical depletion and continued reoxygenation. Bacteria feed on organic matter in an oxidation process which consumes DO, and when there is an ample oxygen and food supply, bacteria multiply rapidly. This bacterial proliferation is essential, for bacteria are an integral part of the food chain which provides nourishment for fish and other higher forms of life. However, other

parts of the chain also consume oxygen, which must therefore be replaced by the twin mechanisms of aeration and photosynthesis if the water is to continue to sustain life. In rapidly moving streams, aeration is generally the more important of the two regenerative processes, but in quiescent lakes and ponds, photosynthesis becomes predominant. With aeration of moving waters there is usually little diurnal variation of DO, but when photosynthesis is the main method of replacing oxygen, there is considerable variation since oxygen production is high during daylight hours while at night there is no production. It is important to know the normal oxygen profile so that deviations from the normal will be readily apparent. Natural fluctuations in DO can be drastically modified by pollution, even leading to the complete disappearance of the DO in a relatively short space of time. If waste is discharged into a stream, it lowers the DO level. When the waste load is not too heavy, the DO concentration sags and then recovers. However, when the waste load is great, then the DO supply may be exhausted, degradation of the aqueous environment begins by anaerobic processes and the water becomes offensively brackish and septic. For most species of fish, the concentration of DO should not fall below 5 mg/L. The normal amount of DO in the cleanest water is extremely small. The concentration of oxygen in water at 20°C and standard pressure is 9.2 mg/L or 9.2 parts per million (ppm).² There is an inverse temperature / oxygen solubility relationship as shown in Figure 1.1, i.e. the concentration of oxygen increases as the temperature of the water drops,² so that the net amount of oxygen present in a river may be some 50% greater in winter than in summer.

In sewage treatment plants, biochemical breakdown of the sewage is achieved by bacterial attack in the presence of oxygen. Failure to maintain an adequate supply of air in these processes leads to increased activity of anaerobic, sulphate-reducing bacteria with the resulting production of hydrogen sulphide; this, in addition to being foul smelling, can cause serious and costly corrosion problems. On the other hand, aeration is the largest single operating expense, and oxygen levels greater than the optimum concentration levels required are wasteful and inefficient. Therefore, DO monitoring is important, and sewage disposal and sanitation engineers need to have a continuous record of the oxygen level at each stage of the treatment process.

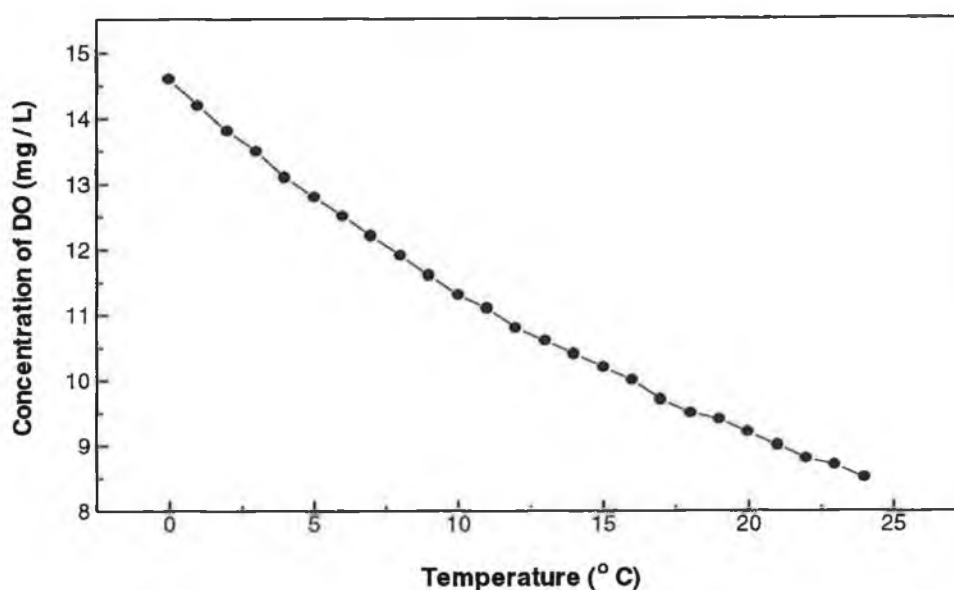


Figure 1.1: Variation of oxygen concentration in water as a function of temperature.

As stated earlier, oxygen is vital for animal and plant life. However, apart from aquatic species, oxygen for life is usually thought of as being needed in gaseous form. But, before a living system can make use of oxygen it must first be dissolved. In the body, whether human or otherwise, oxygen is transported to the cells and tissues by the blood, and clearly a knowledge of DO distribution in blood at various parts of the body can be extremely important for physiological and other medical studies, as can DO monitoring on tissue.

In emphasising the important role of plants as photosynthetic agents, supplying and maintaining the oxygen balance in the environment, it must not be forgotten that they need to breathe as well. Plant respiration has also to be studied in detail. Most plant respiration occurs below ground, thus it is essential that the soil be adequately aerated. Without sufficient aeration, a marked decrease in root activity is found, with mineral salt uptake being affected and the plant ultimately becoming diseased.

Monitoring of oxygen is also very important in the production and preparation of foodstuffs. Fermentation processes are an obvious example, but perhaps less well known is the need for control in processed food preparation to avoid discolourations, in fruit ripening to obtain an optimum rate of ripening, and in meat curing to produce a desirable flavour.

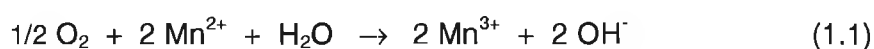
Oxidative corrosion of boiler pipes in steam generators and heaters can occur due

to the high temperatures involved. Even minute traces of DO, of the order of ppb or less, can be extremely detrimental. Hence rigorous control and monitoring must be maintained to prevent the DO level building up.

As can be seen, dissolved oxygen monitoring is extremely important in almost every facet of plant and animal life, demonstrating the need for low-cost, reliable methods of measuring dissolved oxygen, which can be adapted to suit all the above needs.

1.3 Dissolved oxygen monitoring

There are two major techniques employed to measure dissolved oxygen. The first is the Winkler or iodometric method, and the second is the amperometric or electrochemical method. The Winkler method is a titrimetric procedure based on the oxidising property of DO. This method is the most precise and reliable titrimetric procedure for DO analysis. It is based on the addition of divalent manganese solution, followed by strong alkali, to the sample in a glass, stoppered bottle. DO rapidly oxidises an equivalent amount of the dispersed divalent manganous hydroxide precipitate to hydroxides of higher valency states.³ In the presence of iodide ions in acidic solution, the oxidised manganese reverts to the divalent state, with the liberation of iodine equivalent to the original DO content. The iodine is then titrated with a standard solution of thiosulphate. The reaction details are shown in Equations 1.1, 1.2 and 1.3.⁴



Thus one mole of oxygen is equivalent to four moles of thiosulphate. The titration end-point can be detected visually with a starch indicator, or electrometrically, with potentiometric or dead-stop techniques. Experienced analysts can maintain a precision of $\pm 50 \mu\text{g/L}$ with visual end-point detection, and a precision of $\pm 5 \mu\text{g/L}$ with electrometric end-point detection, by measuring a change in conductivity in the solution.

However, this method is not very efficient for DO monitoring due to the difficulty in collection of the samples for measurement and retention of the original DO concentration so that it does not absorb any from the atmosphere. Large sample volumes are required for this method and it is very time consuming.

The second method is the electrochemical or amperometric method, and is based on the reduction of oxygen at an electrode after diffusion across an oxygen permeable membrane. It was first developed by L. C. Clark in 1956,⁵ and consisted of a two-electrode voltammetric cell using a polyethylene membrane. The potassium chloride electrolyte was retained in the cell by the membrane and the material of the membrane was chosen so that oxygen molecules would diffuse through it easily, while acting as a barrier to molecules of other substances. The electrode materials used were silver for the cathode and lead for the anode. Clark cells are today the most common device used to detect and measure dissolved oxygen. Figure 1.2 shows a schematic diagram of a typical Clark electrode.

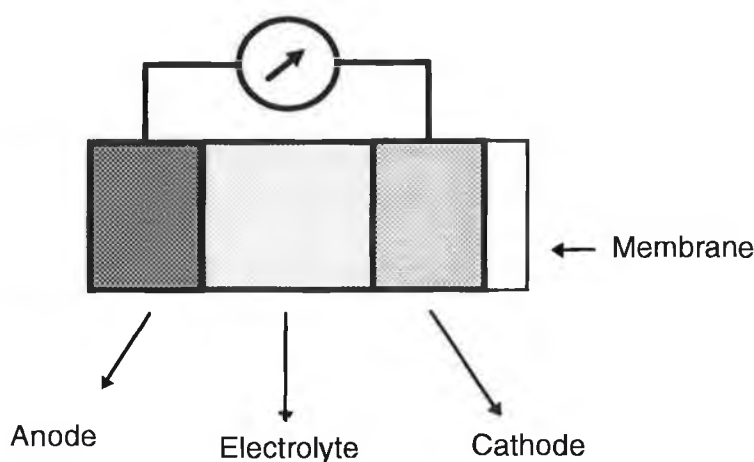
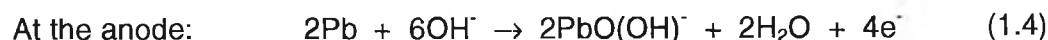


Figure 1.2: Schematic of a typical Clark electrode.

Oxygen diffuses through the porous membrane and is reduced at the silver cathode. This results in an electrical current being generated, the magnitude of which is determined by the rate of diffusion to the lead electrode which is proportional to the partial pressure of oxygen outside the membrane. Thus the current serves as a measure of the partial pressure of oxygen. The half reactions are shown in Equations 1.3 and 1.4.



Diffusion of oxygen through the membrane continues until steady state is reached. The current produced is proportional to the rate of consumption of oxygen, and the oxygen permeability of the diffusion barrier. The response time of the sensor is limited by the diffusion barrier. However, there are problems associated with this type of sensor: ^{6, 7}

- (i) Oxygen is consumed during detection and this can have undesirable effects when the flow of oxygen through the membrane has to be balanced against the rate of electrochemical reduction.
- (ii) The cells have limited lifetimes and require replacement when the platinum cathode has been totally oxidised. The electrolyte is also consumed. Long-term use results in gradual drift in the electrode signal due to electrolyte consumption.
- (iii) Since the electrode actually consumes oxygen, measurements in a stagnant sample cause the oxygen tension at the membrane sample interface to deplete. Therefore controlling the sample renewal rate or flow rate at the membrane will affect the accuracy of the reading with increasing membrane thickness.
- (iv) Acid gases such as CO_2 and SO_2 can be slightly absorbed by the electrolyte and tend to increase the flux of oxygen to the electrode. This results in an enhanced output signal.

Because of these problems which are associated with electrochemical sensors, an alternative method of sensing oxygen merits investigation. Optical sensing based on light variation as a function of measurand is proposed and has a number of advantages over established sensing methods.

1.4 Optical sensors

An optical sensor is one which uses light and changes in light to detect certain analytes or changes in the environment. The terms 'optrode', from *optical electrode*, and 'optode', from the Greek "the optical way", have been used to label

optical chemical sensors.⁸ Depending on the origin of the optical signal, the devices are classified as absorbance (or reflectance), fluorescence, phosphorescence, Raman or infrared sensors. Dramatic progress has been made in this area in the last twenty years due to the advances in optical technology. Fluorescence-based sensing is far superior to absorbance-based sensing because of its higher sensitivity, selectivity and compatibility with laser excitation sources. Numerous organic, bioorganic and inorganic analytes have been shown to be fluorescent under irradiation or to be determinable via a fluorogenic reaction. Miniature light sources, e.g. semiconductor lasers and LEDs, have become available at low prices, as have sensitive photodetectors, which allows their use in simple and safe instrumentation. Optical fibre is readily available which makes possible the transmission of optical signals over large distances. Depending on the field of application, optical sensing can offer some of the following advantages over other sensor types:^{7, 8}

- (i) Ease of miniaturisation allows the development of very small, light, portable optical sensors.
- (ii) Because the primary signal is optical, it is not subject to electrical interferences, thus eliminating the danger of spark induction.
- (iii) Analyses can be performed in real-time since no sampling, with its inherent drawbacks, is necessary.
- (iv) The use of optical fibres in optical sensors offers the possibility of remote/distributed sensing.
- (v) Optical fibre can transmit much more information than an electrical lead and can be used to assay several analytes at the same time.
- (vi) Optical sensors are simple in design - it is inexpensive to replace or substitute components, thus offering the possibility of disposable sensors.

However, optical sensors do exhibit the following disadvantages:

- (i) They can be prone to dye photobleaching and leaching leading to instability in output signal.
- (ii) Ambient light can interfere if lock-in detection and suitable filters are not used.

- (iii) There is a lack of suitable light sources particularly in the uv and ir regions.
- (iv) There is a lack of selective indicators for various analytes of interest.

Despite these limitations, optical sensors are becoming an attractive alternative means of sensing.

To date there has been much work done and many papers published on optical oxygen sensing. One of the first sensors for oxygen was developed by Bergman⁹ in 1968. His work was based on fluorescence quenching by molecular oxygen of fluoranthene immobilised in a porous 'Vycor' glass. Oxygen quenching of fluorescence of dyes on an inorganic adsorbent was first observed and published by Kautsky¹⁰ in the 1930's. Here he explains the mechanism of fluorescence quenching by oxygen. The dye complex is excited by light of a particular wavelength. While in its excited state, oxygen collides with it and forms a charge transfer complex, whereby the energy is transferred to oxygen and non-radiative decay occurs. Hence, no oxygen is consumed in this reaction as it is fully reversible, and the quenching of fluorescence can then be correlated with oxygen concentration. For fluorescence compounds with long excited-state lifetimes,¹¹ the probability of a collision that will result in quenching is much higher than for one with a short lifetime. Thus, this is an important factor when choosing an indicator. Luminescent transition metal complexes, especially ruthenium poly(pyridyl) compounds, are particularly attractive as oxygen sensing species¹² due to their highly emissive metal-to-ligand charge transfer state (MLCT), long fluorescence lifetime (in the order of hundreds of nanoseconds to tens of microseconds), large Stokes shift¹¹, high quenching efficiencies and strong absorption in the blue-green region of the spectrum. These complexes have been extensively studied by Demas *et al.*¹³, with regard to their photophysical and photochemical properties, and these will be discussed further in Chapter 2.

Reagent-mediated optical chemical sensors (optodes) generally require immobilisation of the appropriate reagent on or near an optical fibre or waveguide, in one of a range of possible configurations. The majority of commercially available sensors, e.g. optical oxygen sensor by Joanneum Research, Graz, Austria,¹⁴ have their indicator phase immobilised in a polymer membrane. It has been found, however, that polymers have a number of disadvantages which affect their suitability as an immobilisation phase for optical sensors.¹⁵ Polymeric

networks are less rigid than inorganic glass, which enables rotational and even translational motion. Hence, they may interact not only with the doping molecule but also with external (oxygen) and internal (indicators, monomers, solvent) impurities and quenchers. Polymers have a degradation problem when exposed to environments containing organic solvents or temperatures above 60°C and they also have serious deformation problems after swelling. Hence the investigation of other methods of immobilisation, in particular, entrapment in sol-gel glasses was considered important.

1.5 The sol-gel process and sol-gel sensors

The development of inorganic host materials with extended chemical and photochemical stability and good optical properties for immobilisation of organic dyes for sensor applications has been the subject of much research during the past few years. One of the most promising approaches has been to use the sol-gel process for the encapsulation of optically-active compounds into a silica glass matrix. Compared with polymers, glass provides the properties which are desired as a sensor substrate such as high surface area, due to the porous nature of the structure.¹⁶ Sensor preparation is simple and avoids the complex chemistry often associated with other immobilisation methods. In this work, doped sols are used to coat planar and fibre substrates forming thin film coatings. The coating produced is tough, inert, intrinsically bound to the substrate, and is considerably more resistant than, for example, polymer films in aggressive environments; the coatings have high chemical, photochemical and thermal stability, and have low optical absorption in the visible and near infrared region. All these factors along with low fabrication temperature, contribute to the suitability of porous sol-gel films as an immobilisation phase for optical chemical sensing. The sol-gel process is being used increasingly in the fabrication of chemical and biochemical sensors. Generally, sensor molecules are entrapped in the sol-gel-derived cage-like microstructure, which can still be permeated by smaller analyte molecules. Monolithic or thin-film structures may be fabricated, although the latter offer the advantage of faster response time. The films are fabricated from silicon alkoxide precursors, which undergo hydrolysis and polycondensation reactions followed by a temperature programme which controls the densification process. If the temperature is limited to less than 200°C, a non-densified porous glass matrix is

formed. This microporous glass acts as a support matrix for analyte-sensitive dyes which are added to the silicon alkoxide solution. The dye molecules are entrapped in the nanometre-scale cages formed by the cross-linking silicon and oxygen units. Smaller analyte molecules can permeate the matrix and access the dye complex in the pores. By selecting fabrication parameters appropriate to the size of the dopant molecule, leaching is negligible. Variation of the processing parameters allows tailoring of the microstructure of the films for different sensing applications. The pioneers in the field of sol-gel-coated optical fibre chemical sensors were MacCraith *et al.*¹⁷ and Ding *et al.*¹⁸ MacCraith developed a pH optrode, based on coating an unclad silica fibre with a porous silica cladding within which fluorescein, a pH-sensitive dye, was entrapped. The sol-gel fluorescent cladding was excited by the evanescent wave of argon-ion laser radiation ($\lambda_{\text{ex}} = 488 \text{ nm}$) guided by the sensing fibre. The resulting fluorescence ($\lambda_{\text{em}} = 530 \text{ nm}$) was detected and recorded as a function of pH. A different approach was taken by Ding *et al.* In their work, the pH sensor was prepared by co-immobilising absorption-type pH indicators into silica gel, which was coated as a thin film into a porous core silica glass fibre. In this sensor, the analyte penetrates into the porous fibre core and interacts with the pH indicator where on-line absorption takes place. Since then, sol-gel processing has been widely used for optical chemical sensors and biosensors in the detection of numerous analytes e.g. ammonia¹⁹, nitrogen monoxide²⁰, carbon monoxide²⁰, hydrogen sulphide²¹, and oxygen¹², to name but a few. The analyte of interest in this project is oxygen, initially in the gas phase for preliminary work, but primarily in the dissolved phase for use in various applications already mentioned in Section 1.2.

1.6 Outline of thesis and objectives of the project

This thesis describes the development of an optical dissolved oxygen sensor. The first three chapters provide background information on dissolved oxygen sensing, fluorescence quenching and the sol-gel process. Chapters 4 and 5 describe the experimental technology involved in sensor fabrication and characterisation. Chapters 6, 7 and 8 contain experimental results and analysis, while Chapter 9 summarises the work and discusses future developments arising from the project.

The following are the list of objectives at the start of the project:

(A.) Preliminary investigation of ruthenium-doped sol-gel glass as dissolved oxygen sensor.

This involved laser-based intensity and lifetime measurements in gas and aqueous phase.

(B.) Design and construction of dissolved oxygen characterisation system.

There were a number of strands involved in this section:

- (i) Design of sealed sample compartment with optical access ensuring good sample mechanical stability.
- (ii) Design and assembly of LED driving circuitry and photodiode detection circuitry to enable construction of compact sensor, and interfacing these electronics to a PC for control and signal processing
- (iii) Assembly of optics which involves choosing appropriate LED, photodiode, filter and lenses to ensure efficient excitation of the sample and efficient collection of fluorescence.
- (iv) Construction of gas mixing and circulation system in order to achieve water of stable DO concentrations.

(C.) Material characterisation and optimisation.

Having identified that ruthenium-doped sol-gel films responded to dissolved oxygen, it was then necessary to improve on and optimise the films for this application. This would involve:

- (i) Improving the quenching response of the films in DO.
- (ii) Investigation of initial pH conditions on the sol-gel film.
- (iii) Investigation of heat treatment for optimisation of oxygen accessibility.
- (iv) Ensuring film stability in water i.e. no leaching or film degradation.
- (v) Optimisation of the dopant concentration for DO sensing.

(D.) Investigation of the fundamental quenching process in materials

This would involve investigation of the oxygen diffusion behaviour and accessibility of the dye complex

(E.) Optimisation of ruthenium-doped sol-gel coating.

Having found the optimum film for use as a DO sensor, a number of tests must be performed to ensure the stability and long-life of the sensor.

- (i) Assessment of long-term mechanical stability of coating.
- (ii) Assessment of long-term quenching response.
- (iii) Investigation of the effects of temperature variations on the coating.
- (iv) Investigation of possible interferents on sensor performance.
- (v) Investigation of long-term dye leaching.

(F.) Development of a portable version of the sensor for preliminary field testing.

The above objectives will be revisited in Chapter 9 with respect to the level of achievement of these goals.

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CHAPTER 2:

OXYGEN SENSING BY FLUORESCENCE QUENCHING

2.1 Introduction to fluorescence

Fluorescence occurs when an electron in an atom or molecule, having been excited from a ground state energy level (absorption process) to a high energy level, de-excites to the ground state resulting in the emission of a photon of light. All sensors in this work involve fluorescence emission.

Figure 2.1 shows a simplified energy level diagram which illustrates the processes of absorption and emission.

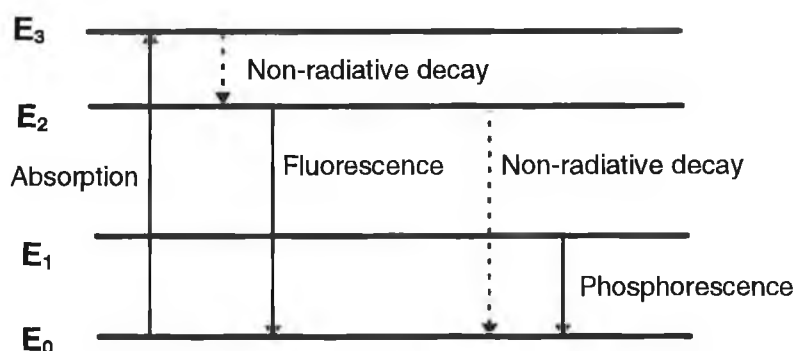


Figure 2.1: Generalised energy level diagram illustrating the fundamental excitation and emission processes.

Each electron in a molecule carries a spin angular momentum with a spin quantum number, $s = 1/2$. The electron possesses a 'spin-up' or 'spin-down' property which is indicated by the use of arrows $\uparrow$ or $\downarrow$, or the symbols, α and β . The total spin angular momentum possessed by a many-electron atom or molecule is represented by the total spin quantum number, S , which may be calculated as the vector sum of all the individual contributions from each electron.¹ Two electrons each possessing $s = 1/2$ may be present with their spins parallel or opposed. If the spins are opposed the total spin quantum number, S , is zero. If the electron spins are parallel, the total quantum number, S is $1/2 + 1/2 = 1$. The spin multiplicity gives the number of states expected in the presence of an applied magnetic field and is given by $2S + 1$. Thus a molecule with all electrons spin-paired possesses $S = 0$ and a spin multiplicity of 1. Such an electronic state is

referred to as a singlet state. The combination of ground state and singlet state is abbreviated by the symbol S_0 . This corresponds to E_0 in the generalised diagram shown in Fig. 2.1. The interaction of a molecule with light leads to excitation whereby an electron is promoted to an excited state. The excited state can either be a singlet or S state for example E_2 in Fig. 2.1 or a triplet or T state where the spin quantum number is 1, giving a spin multiplicity of 3. This excited state is represented by E_1 in Fig. 2.1. If a molecule with a ground state energy level, E_0 , is excited by an incoming photon, it can be raised to an excited state, E_3 . The absorbed energy can be released either radiatively or non-radiatively. If the gap between E_3 and E_2 is small, the electron in that excited state tends to decay non-radiatively by phonon emission, releasing energy as heat to the material. Radiative decay occurs either from the singlet (E_2) or the triplet state (E_1). Fluorescence is the emission resulting from the return of the electron to the ground state, if this transition originates in a singlet state. Such transitions are quantum mechanically allowed, with emission rates as high as 10^8 per second, and consequently fluorescence lifetimes of approximately 10 ns.² Emission can also occur when a triplet state excited electron (from E_1) returns to a singlet ground state (E_0). This is known as phosphorescence. These transitions between states of different multiplicity are quantum mechanically forbidden, but in some molecules are weakly allowed. This happens when the molecule contains a heavy atom, because its strong spin-orbit interaction can reverse the relative orientations of pairs of electrons. Emission rates are slow, and hence fluorescence lifetimes are longer. The fluorescence lifetime is the average period of time a fluorophore remains in the excited state prior to its return to the ground state.³ The observed lifetime depends on the radiative and non-radiative decay rates as shown by Equation 2.1,

$$k_{obs} = \frac{1}{\tau_{obs}} = \frac{1}{\tau_{rad}} + \frac{1}{\tau_{non-rad}} \quad (2.1)$$

where $1/\tau$ is equal to the decay rate, k and where obs., rad., and non-rad., refer to the observed, radiative and non-radiative lifetimes, respectively. The fluorescence complex of interest in this work has a singlet ground state and a triplet emitting state resulting in a partially allowed transition of relatively long fluorescence lifetime. This is discussed in the next section.

2.2 Transition metal complexes for O₂/DO sensing

In the past, organic dyes such as pyrene have been used as fluorescent indicators in the detection of oxygen. These indicators, however, suffer from the disadvantage of having both short-wave excitation and small Stokes shift, i.e. the difference in wavelength between the absorption and fluorescence maxima of the indicator complex, and both of these add complexity to the measurement instrumentation. Fluorescent transition metal complexes, especially ruthenium poly(pyridyl) compounds, have become increasingly important as oxygen sensing species.⁴ These complexes have very desirable features.² They exhibit strong absorption in the blue-green region of the spectrum, for which a range of suitable laser and LED sources is available. They have a relatively large Stokes shift, and this enables the use of filters, which serve to minimise the detection of excitation light with minimal reduction of the fluorescence signal, and this in turn simplifies sensor design and reduces costs. Long unquenched excited-state lifetimes result in high sensitivity according to the Stern-Volmer equations (see Section 2.3). A further consequence of this is that relatively inexpensive sensing instrumentation is required for lifetime-based sensors. These compounds also tend to be thermally, chemically and photochemically robust, all of which add stability to the sensor and extend sensor lifetime.

Transition metal complexes are characterised by partially filled d orbitals.^{2, 5} To a considerable extent, the ordering and occupancy of these orbitals determine emissive properties. The ruthenium complex used in this sensor is an octahedral MX₆ d⁶ metal complex, where M is the metal and X is the ligand. An orbital and state diagram representative of this complex is shown in Figure 2.2. The octahedral field of the ligand splits the five degenerate d orbitals by an amount Δ , into a triply degenerate t_{2g} level and a doubly degenerate e_g level. The t_{2g} orbitals point in between the x, y and z axes, and the e_g orbitals point along the axes. The splitting arises because the two e_g orbitals are directed toward the six ligands and the remaining t_{2g} orbitals point between the ligands,⁵ resulting in the e_g orbital being higher in energy than the t_{2g} orbital.

The magnitude of the splitting is given by the crystal field splitting, Δ , which is a particularly important parameter whose size is determined by the crystal field strength of the ligands and the central metal ion.⁶ The fluorescence properties of the complex can be controlled by altering the ligand, geometry and metal ion. The distribution of electrons between the t_{2g} and e_g levels is greatly affected by Δ . If Δ

is large, i.e. a strong field, it is energetically more favourable to pair electrons in the t_{2g} level, than to keep them unpaired by distributing them throughout the t_{2g} and e_g levels. Here only strong crystal field systems are considered, such that all six d electrons pair and fill the three t_{2g} orbitals.

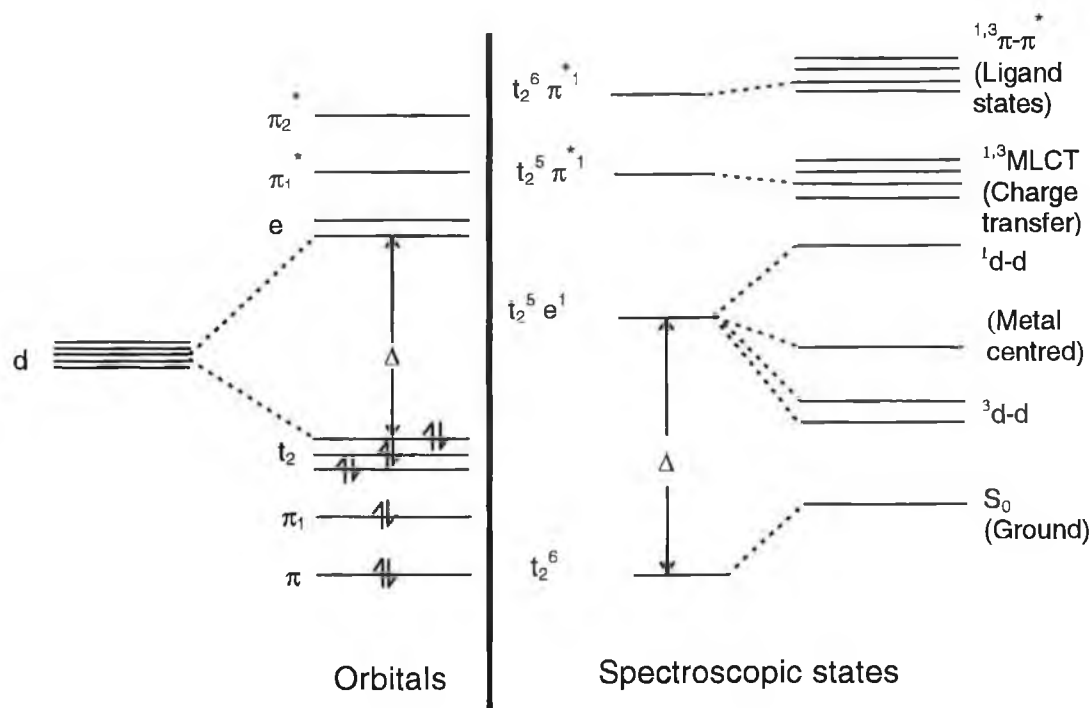


Figure 2.2: Simplified orbital and state diagrams for a $MX_6 d^6$ metal in an octahedral environment showing d and π bonding and π^* antibonding orbitals.²

The ligands have π and σ orbitals, but only the π orbitals are spectroscopically important for visible and near-uv absorptions and emissions. There are both π bonding and π antibonding (π^*) levels, and the π bonding levels are filled. Because all spins are paired, the ground state is a singlet, as discussed in the previous section. The lowest excited states are derived from promoting an electron to one of the unoccupied orbitals. There are three types of excited states which are of spectroscopic importance in an octahedral field. The first is where a d electron is promoted to another d level, i.e. a d-d transition. The second is a $\pi-\pi^*$ state, where an electron from a π bonding orbital is promoted to a π^* antibonding orbital. The third is where a d electron can be promoted to a π^* antibonding orbital, or an electron in a π bonding orbital can be promoted to an unfilled d orbital. These

states are known respectively as metal to ligand charge transfer states (MLCT) and ligand to metal charge transfer states (LMCT).

The lowest ligand excited states are $\pi\text{-}\pi^*$ states derived from promoting a bonding π electron to a π^* level. There are triplets and singlets, and the triplet is always below its analogous singlet state. These transitions are largely localised on the organic ligands and are spectroscopically similar to those of the free ligand. In these complexes, charge transfer transitions are more strongly allowed than d-d transitions, hence have shorter radiative lifetimes and are less susceptible to intramolecular and environmental quenching. The molar extinction coefficients of the spin-allowed transitions are large, and as a result are easier to pump optically.

2.2.1 Photochemistry of O_2 / DO sensing ruthenium complexes

The photophysical and photochemical properties of ruthenium(II) polypyridyl complexes have been studied extensively and have attracted a lot of attention due to the unique combination of ground- and excited-state properties.

Ru(II) compounds are stable d^6 species, which can be oxidised (removal of a metal localised electron), or reduced (addition of an electron into a ligand π^* orbital). The control of the fluorescence properties of these complexes is dependent on the relative state energies and the nature and energy of the lowest excited state.⁷ The positions of the lowest d-d, MLCT and $\pi\text{-}\pi^*$ triplet states are shown in Figure 2.3.

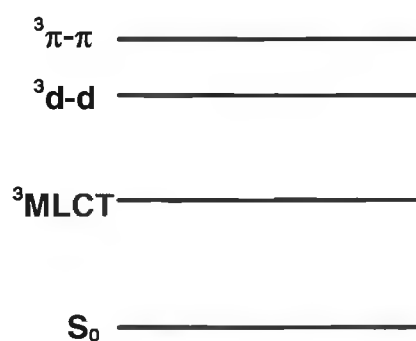


Figure 2.3: Lowest triplet orderings for ruthenium metal complexes $[\text{Ru(II)L}_3]^{2+}$ showing d-d, $\pi\text{-}\pi^*$ and MLCT states.

The metal to ligand charge transfer transition is the lowest in energy with respect to all the other possible excited state transitions. As a consequence, it is this state

that is mainly responsible for the absorption and emission characteristics of the complex. The long lifetimes of these states cause the high susceptibility to oxygen quenching. Any d-d states must be well above the emitting level (MLCT) to prevent their thermal excitation. It is also important to note that the emitting state cannot be too low in energy, as the energy gap law states that the radiationless processes become more efficient as the emitting state approaches the ground state.²

$[\text{Ru(II)L}_3]^{2+}$ is the general form of the ruthenium complexes, where L denotes the ligands, which are primarily α -diimine ligands. The prototype of these Ru(II) complexes is $[\text{Ru}^{\text{II}}(\text{bpy})_3]^{2+}$, ruthenium-tris (2, 2-bipyridine) which is an oxygen-sensitive species. Other important examples are ruthenium tris-1,10-phenanthroline, $[\text{Ru}^{\text{II}}(\text{phen})_3]^{2+}$ and ruthenium-tris(4,7-diphenyl-1,10-phenanthroline), $[\text{Ru}^{\text{II}}(\text{Ph}_2\text{phen})_3]^{2+}$. The complex used in this work is the latter complex, $[\text{Ru}^{\text{II}}(\text{Ph}_2\text{phen})_3]^{2+}$ due to its longer lifetime and larger size compared with the other three complexes, and also the fact that it is largely unaffected by changes in the microenvironment.⁷ These factors make the complex more suitable for quenching by oxygen.

The $[\text{Ru}^{\text{II}}(\text{Ph}_2\text{phen})_3]^{2+}$ has its absorption maximum at 450 nm, and this is assigned to the $^1\text{MLCT}$ transition. Other lesser absorption peaks occur at 417, 263 and 224 nm.⁸ The fluorescence maximum in aqueous solution occurs at 608 nm, and is assigned to the Stokes-shifted emission of the $^3\text{MLCT}$ excited state to the ground state. The excited state transitions of $[\text{Ru}^{\text{II}}(\text{Ph}_2\text{phen})_3]^{2+}$ are shown in Fig. 2.4.

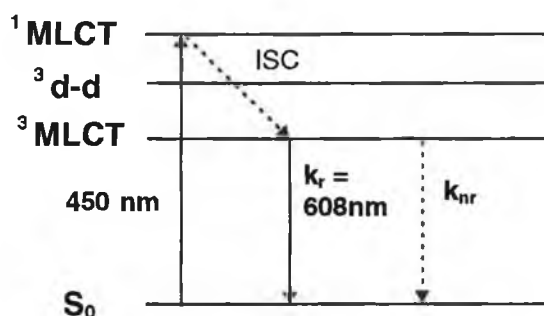


Figure 2.4: Excited state transitions of $[\text{Ru}^{\text{II}}(\text{Ph}_2\text{phen})_3]^{2+}$.

The fluorescence quantum yield for this complex is 0.5, and it has a molar extinction coefficient of 30,000 at 450 nm.⁹ Non-radiative decay by intersystem crossing (ISC) can occur from the singlet MLCT state to the triplet MLCT state.

Fluorescence emission is characterised by the radiative decay, k_r , of this excited $^3\text{MLCT}$ state. According to Xu et al,⁹ $[\text{Ru}^{(\text{II})}(\text{Ph}_2\text{phen})_3]^{2+}$ has one of the highest responses to oxygen that they have observed.

2.3 Quenching of fluorescence and Stern-Volmer equation

Fluorescence quenching refers to any process which decreases the fluorescence intensity of a given substance. A variety of processes can result in quenching. These include excited state reactions, energy transfer, complex formation, and collisional quenching.³ In this work, we are concerned primarily with quenching resulting from collisional encounters between the fluorophore and the quencher, which is called collisional or dynamic quenching. Static quenching will also be mentioned, and this is due to complex formation.

Both static and dynamic quenching require molecular contact between the fluorophore and quencher. In the case of collisional quenching, which is illustrated in Figure 2.5, the quencher must diffuse to the fluorophore during the lifetime of the excited state. Upon contact, the fluorophore returns to the ground state, without emission of a photon. In Figure 2.5, S_0 , S_1 and S_2 denote the singlet ground state, the triplet excited state and the singlet excited state.

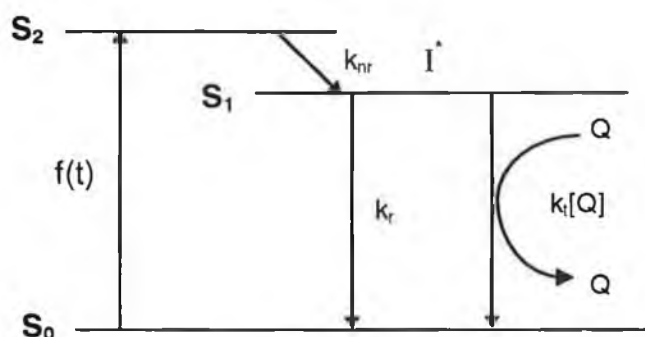


Figure 2.5: State diagram for collisional quenching.²

Absorption occurs from S_0 to S_2 , and intersystem crossing results in non-radiative decay from S_2 to S_1 , the triplet state. It is at this stage that quenching occurs. The quencher collides with the excited complex, and the complex then decays non-radiatively with decay rate, $k_t[Q]$. According to Equation 2.1, the observed decay is composed of both radiative and non-radiative decay. As the concentration of quencher increases, the non-radiative decay rate increases, and thus the

observed lifetime will decrease with accompanying decrease in fluorescence intensity.

Collisional quenching of fluorescence is described by the Stern-Volmer equation,

$$\frac{I_0}{I} = 1 + k \tau_0 [Q] = 1 + K_{SV} [Q] \quad (2.2)$$

$$\frac{\tau_0}{\tau} = 1 + k \tau_0 [Q] = 1 + K_{SV} [Q] \quad (2.3)$$

In this equation, I_0 and I are the fluorescence intensities in the absence and presence of quencher, respectively, $[Q]$ is the concentration of quencher, τ_0 and τ are the lifetimes of the fluorophore in the absence and presence of quencher, respectively, and K_{SV} is the Stern-Volmer quenching constant which is defined as follows:

$$K_{SV} = k \tau_0 \quad (2.4)$$

where k is the bimolecular quenching constant which is defined to be

$$k \propto \alpha D \quad (2.5)$$

where α is the oxygen solubility in the matrix and D is the diffusion coefficient of oxygen in the sensing film. Quenching data are often presented as a plot of I_0/I versus $[Q]$, as I_0/I is expected to be linearly dependent on the quencher concentration. The Stern-Volmer plot yields an intercept of 1 on the y-axis and a slope of K_{SV} . A linear Stern-Volmer plot generally indicates that all the fluorophore is equally accessible to the quencher. If two fluorophore populations are present, and one is not accessible to quencher, then the Stern-Volmer plots deviate from linearity towards the x-axis. This is also the case if there are two or more distributions of fluorophore which result in varying accessibility by the quencher. The latter is a more realistic and common situation.

Another type of fluorescence quenching by oxygen exists which is known as static quenching.³ This process occurs as a result of the formation of a non-fluorescent ground state complex between the fluorophore and the quencher. When this complex absorbs light it immediately returns to the ground state without the emission of a photon. The lifetimes or temperature dependence can be used to distinguish between contributions from static and collisional quenching. In the case of static quenching a fraction of the fluorophores is removed from

observation such that a background light level exists. The complexed fluorophores are non-fluorescent and the only observed fluorescence is from the uncomplexed fluorophore. If the τ_0 / τ and I_0 / I plots versus $[Q]$ are linear and coincide, quenching is purely collisional. However, if I_0 / I is everywhere greater than τ_0 / τ , static quenching is present. Therefore from the analysis of both intensity and lifetime quenching data, the determination of the static and dynamic quenching contributions may be carried out. In this work there were no contributions from static quenching.

One of the best known collisional quenchers is molecular oxygen, and in many cases, it is necessary to remove dissolved oxygen in order to obtain reliable measurements of the fluorescence yields or lifetimes. For us this is very encouraging as dissolved oxygen is the analyte which we want to sense, and fluorescence quenching is the sensing mechanism. Oxygen has a triplet ground state and a low-lying singlet excited state that can efficiently deactivate a variety of singlet and triplet emitters. The quenching of luminescence by oxygen was first documented by Kautsky in 1939.¹⁰ In this work he describes the effect of oxygen on the luminescence of dyes on an inorganic adsorbant. In 1919, Stern and Volmer published a paper documenting the variation of intensity with quencher concentration.¹¹ This relation has since become known as the Stern-Volmer equation, and has been used to describe the oxygen quenching process by a number of researchers.^{3, 12, 13}

2.3.1 Stern-Volmer equation

If a pulse of light of a suitable wavelength is directed at a bulk fluorophore, a certain portion of the fluorophore molecules will be excited. The excited molecules will de-excite at a rate which is exponential with a rate constant, K . K is composed of the sum of several rate constants:¹²

$$K = k_r + k_{nr} + k_t[Q] \quad (2.6)$$

where k_r is the radiative decay constant, k_{nr} is the non-radiative decay constant, k_t is the rate constant for the transfer of energy to the quencher, and $[Q]$ is the concentration of quencher molecules in the bulk fluorophore. The fluorescence quantum yield, η , is defined for a particular fluorophore as the ratio of the number of photons emitted to the number of photons absorbed, and can be written in terms of radiative and non-radiative decay constants as follows:

$$\eta = \frac{k_r}{k_r + k_{nr}} \quad (2.7)$$

Hence it can be shown that the fluorescence quantum yield, η , in the presence of a quencher, is given by

$$\eta = \frac{k_r}{k_r + k_{nr} + k_t [Q]} \quad (2.8)$$

In the case where the only available de-excitation process is fluorescence, k_t and k_{nr} are equal to zero, hence $\eta=1$. As the other de-excitation processes, k_{nr} and k_t become available, η diminishes. Therefore, the ratio of η_0 , the fluorescence quantum yield in the absence of quencher, to η , the fluorescence quantum yield in the presence of a quencher concentration $[Q]$ can be expressed as:

$$\frac{\eta_0}{\eta} = \frac{\left(\frac{k_r}{k_{nr} + k_r}\right)}{\left(\frac{k_r}{k_r + k_{nr} + k_t [Q]}\right)} = 1 + \left(\frac{1}{k_r + k_{nr}}\right) k_t [Q] \quad (2.9)$$

Hence

$$\frac{\eta_0}{\eta} = 1 + \tau_0 k_t [Q] \quad (2.10)$$

Then using the fact that

$$\frac{\eta_0}{\eta} = \frac{I_0}{I} \quad (2.11)$$

where I_0 and I are the fluorescence intensity in the absence and presence of quencher, we obtain the Stern-Volmer expression

$$\frac{I_0}{I} = 1 + K_{SV} [Q] \quad (2.12)$$

where $K_{SV} (= k_t \tau_0)$ is the Stern-Volmer constant.

In the case of dynamic quenching, $I_0/I = \tau_0/\tau$, therefore

$$\frac{\tau_0}{\tau} = 1 + K_{SV} [Q] \quad (2.13)$$

Equations 2.12 and 2.13 describe a linear relationship between change in fluorescence intensity / lifetime with quencher concentration i.e. the fluorophores are fully and equally accessible to the quencher.

2.3.2 Concentration quenching

In order for fluorescence to be observed, absorption must occur. Absorption can cause many problems during a fluorimetric assay, just as fluorescence causes a problem when the absorbance of a sample is measured. Equation 2.14 describes the relationship between the fluorescence signal from a fluorophore and dye concentration¹⁴

$$I_f = \eta I_0 [1 - \exp(-\epsilon L c)] \quad (2.14)$$

where I_0 is the intensity of incident radiation, ϵ is the molar extinction coefficient, L is the optical pathlength in the sample, c is the dye concentration, and η is the fluorescence quantum efficiency. For low concentrations ($\epsilon \eta c \ll 1$), the fluorescence intensity is directly proportional to the dye concentration

$$I_f = \eta I_0 \epsilon L c \quad (2.15)$$

Thus, the more highly absorbing the sample, the greater the fluorescence. When the absorption is too large, however, no light can pass through to cause excitation. At higher concentrations, the light is not evenly distributed along the path of light. The layer nearest the light source absorbs much radiation leaving less available for the rest of the sample layers. As a result, considerable excitation occurs throughout the front of the sample, such that the incident intensity is progressively reduced as it passes through the sample. This type of concentration quenching causes a fluorescence loss that is called the inner-cell effect.

As will be seen later, in Section 4.5 on optimisation of the ruthenium complex concentration for DO sensing, concentration quenching is thought to be a factor in the increase and subsequent decrease of the response of the sensor to oxygen with increasing ruthenium concentration.

2.4 Sensing by quenching of fluorescence

Sensing by quenching of fluorescence is an efficient method for detecting and measuring various analytes. This technique involves monitoring the variation of fluorescence intensity i.e. the ratio of unquenched fluorescence to quenched fluorescence, with quencher concentration, according to the Equation 2.12. This can be applied to the oxygen quenching process, and used to measure the concentration of oxygen in a sample.

It is a very popular measurement technique, and has been used by many groups to sense various analytes. Kroneis and Marsoner (1983) documented a

fluorescence-based sterilizable oxygen probe for use in bioreactors.¹⁵ In 1989, Trettnak and Wolfbeis immobilised an oxygen-sensitive dye complex on nylon membranes, and detected the amount of lactate by measuring the changes in fluorescence intensity.¹⁶ Wolfbeis *et al.* described the development of a fluorosensor for carbon dioxide and oxygen in 1988.¹⁷ Furthermore Demas and Bacon,¹⁸ Cox and Dunn¹⁹, and MacCraith *et al.*²⁰ have used the fluorescence intensity technique to detect oxygen in gas phase. This work deals mainly with intensity-based sensing of dissolved oxygen.

2.5 Fluorescence lifetime sensing

Fluorescence lifetime measurements are based on the variation of the fluorescence decay-time with oxygen concentration according to Equation 2.13. This method has a number of advantages over fluorescence intensity measurements, as decay-times are absolute quantities, which offer the possibility of inherent referencing and are usually independent of light source instability, light throughput, detector sensitivity and leaching / photobleaching of dye.²¹

Two methods are widely used for the measurement of fluorescence lifetimes, the pulse method and the harmonic or phase modulation method.³ In the pulse method, the sample is excited with a brief pulse of light and the time-dependent decay of fluorescence intensity is measured. In the phase modulation method, the sample is excited with sinusoidally modulated light. The phase shift and demodulation of the emission, relative to the incident light, is used to calculate the lifetime. The first method which we will consider is the pulse method.

2.5.1 Pulse method

Consider the excitation of a fluorophore with an infinitely short pulse of light, resulting in an initial population, N_0 , of fluorophores in the excited state. The rate of decay of the initially excited population is ³

$$\frac{dN(t)}{dt} = -(k_r + k_{nr})N(t) \quad (2.16)$$

where $N(t)$ is the number of excited molecules at time t following excitation, k_r is the radiative decay rate and k_{nr} is the rate of non-radiative decay. As $N(t) = N_0$ at $t = 0$, integration of Equation 2.16 yields

$$N(t) = N_0 e^{-t/\tau} \quad (2.17)$$

where $\tau = (k_r + k_{nr})^{-1}$ is the observed lifetime of the excited state. Hence, we expect the fluorescence intensity, $I(t)$, which is proportional to the excited state population [$I(t) = k_r N(t)$], to decay exponentially. The fluorescence lifetime is the time required for the intensity to decay to $1/e$ of its initial value, and this time is $(k_r + k_{nr})^{-1}$. Alternatively, the lifetime may be determined from the slope of a plot of the $\log I(t)$ versus t (see Figure 2.6).

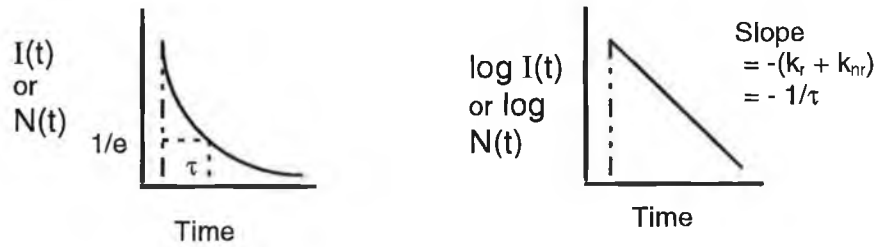


Figure 2.6: Schematic of pulse lifetime measurements.

Equation 2.17 describes the single exponential decay of a fluorophore. However, fluorophores are often exposed to a range of different environments within a sample, in which case the observed decay is fitted to a sum of exponentials

$$I(t) = \sum_i \alpha_i e^{-t/\tau_i} \quad (2.18)$$

where α_i is a pre-exponential factor representing the fractional contribution to the time-resolved decay of the component with a lifetime τ_i . Non-exponential decays are, generally, as a result of several different fluorophores or one fluorophore in several different environments, and will be referred to as heterogeneous samples. For heterogeneous samples, in which each species decays exponentially, the decay can be described by Equation 2.18. The τ_i values represent the lifetimes of the individual species. The fractional intensity, f_i , of each species is given by

$$f_i = \frac{\alpha_i \tau_i}{\sum_i \alpha_i \tau_i} \quad (2.19)$$

Therefore, the time-resolved decays can be used to reveal and analyse heterogeneous emissions.

Chapter 5 details a system used for measuring lifetimes using the pulse method. This system was used for preliminary sample characterisation which indicated the viability of the samples for lifetime sensing.

2.5.2 Phase fluorimetry

The second method by which lifetimes are routinely measured is the phase-modulation method. Instead of square-wave pulsed excitation the sample is excited with light whose intensity is modulated sinusoidally. The emission is a forced response to the excitation, and therefore the emission is modulated at the same angular frequency, ω , ($\omega = 2\pi \cdot \text{frequency}$) as the excitation.²²

According to the Stern-Volmer equation, Equation 2.13, the lifetime of the complex is quenched by oxygen, just as the intensity is, hence the lifetime of the fluorophore reduces with increasing quencher concentration. Because the phase shift is dependent on the lifetime of the fluorescent material, the concentration of oxygen may be deduced by measuring the phase shift of the emission relative to the excitation signal.

Phase fluorimetry involves the use of a continuous, sinusoidally-modulated excitation combined with phase-sensitive detection. The fluorescent species is excited with light having a time-dependent intensity, $E(t)$, of the form³

$$E(t) = B + A \sin \omega t \quad (2.20)$$

where B is the d.c. component of the exciting light, the degree of modulation, M , is defined as A/B , and ω is the angular modulation frequency. The resulting time-dependent emission is modulated at the same frequency as the excitation.²² Because of the finite lifetime of the excited state, the modulated emission is delayed in phase by an angle ϕ , relative to the excitation.²³ Furthermore, the emission is demodulated relative to the excitation, i.e. the relative amplitude of the variable portion of the emission (a/b) is smaller for the emission than for the excitation (A/B). The phase shifted emission, $R(t)$, is written as follows for a single exponential decay

$$R(t) = b + a \sin(\omega t - \phi) \quad (2.21)$$

Figure 2.7 shows that either the phase angle, ϕ , or the demodulation factor, m , [$m = Ba/Ab$] can be measured and used to calculate the fluorescence lifetime, τ .

The following relationships have been shown to exist between phase shift, demodulation factor and modulation frequency²²

$$\phi = \arctan(\omega \tau) \quad (2.22)$$

$$m = [1 + \omega^2 \tau^2]^{-1/2} \quad (2.23)$$

The above equations apply to a single exponential decay, but in this work the

decays are multi-exponential and need a more rigorous description. The multi-exponential phase fluorimetric versions of Equations 2.22 and 2.23 are given as follows:²⁴

$$\tan \phi = \frac{\sum_{i=1}^n f_i \sin \phi_i \cos \phi_i}{\sum_{i=1}^n f_i \cos^2 \phi_i} \quad (2.24)$$

where ϕ_i is the phase angle associated with the decay time, τ_i , and f_i is the fractional intensity of each lifetime component and is related to the pre-exponential factor, α_i , by Equation 2.19. The resultant demodulation factor, $\langle m \rangle$, is given by

$$\langle m \rangle = \left[\left(\sum_{i=1}^n f_i \sin \phi_i \cos \phi_i \right)^2 + \left(\sum_{i=1}^n f_i \cos^2 \phi_i \right)^2 \right]^{1/2} \quad (2.25)$$

If the degree of modulation or the phase shift are known, it is possible to deduce the lifetime using Equation 2.25. Thus, knowing τ and τ_0 it is possible to calculate the oxygen concentration for a given film (i.e. for a particular K_{SV}) using the Stern-Volmer lifetime equation, Eqn. 2.13. The system used for phase fluorimetric measurements is shown and detailed in Chapter 5.

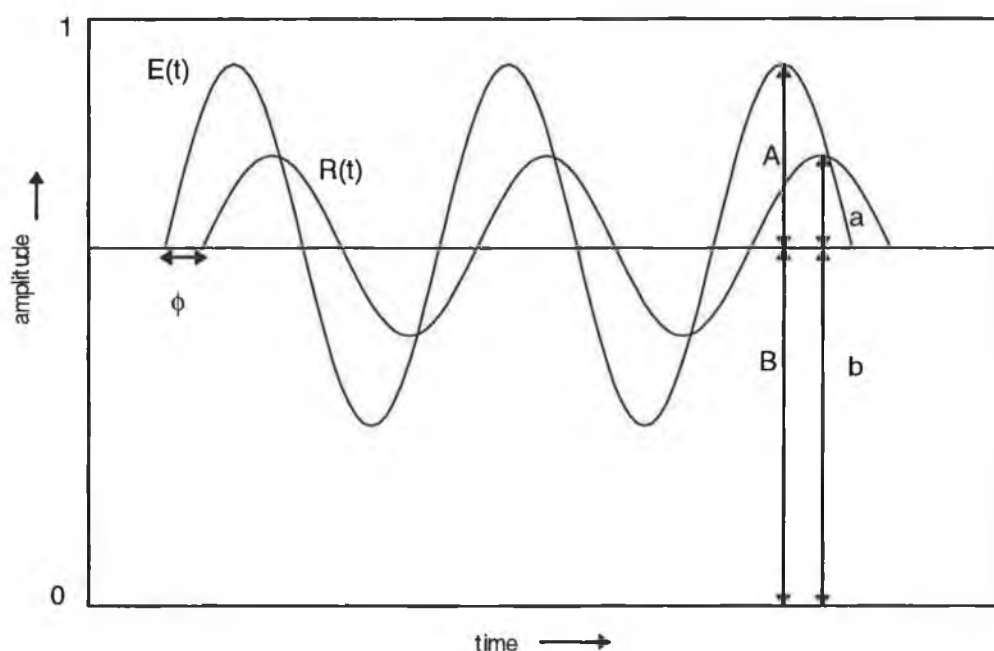


Figure 2.7: Illustration of phase and modulation measurements.

2.6 Conclusions

Transition metal complexes, in particular those of ruthenium, have been described in detail, and the reasons for their suitability as oxygen sensing compounds have been outlined. The various techniques used in this work for sensing oxygen have been illustrated i.e. fluorescence intensity sensing, and the two fluorescence lifetime sensing techniques, the pulse lifetime method and the phase fluorimetric method. Fluorescence lifetime sensing is a more reliable and sensitive method for oxygen detection, and for this particular application, phase fluorimetry is a more suitable technique, as it lends itself more readily to miniaturisation. Both intensity- and lifetime-based sensing are dealt with in Chapters 6, 7 and 8.

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CHAPTER 3: THE SOL-GEL PROCESS

3.1 Introduction

The interest in the sol-gel processing of inorganic ceramic and glass materials began as early as the mid-1800's with Ebelman and Graham's studies on silica gel.¹ Ebelman (1846) prepared the first metal alkoxide from SiCl_4 and alcohol, and found that it gelled on exposure to the atmosphere.² It was finally recognised in the 1930's that alkoxides could be used in the preparation of oxide films, whereupon this process was developed by the Schott glass company in Germany. In 1864, Graham showed that water in silica gel could be exchanged for organic solvents, and hence it was these early investigators who observed that the hydrolysis of tetraethylorthosilicate, under acidic conditions, yielded SiO_2 in the form of a "glass-like material". Fibres could be drawn from the viscous gel and even monolithic optical lenses or composites formed. However, extremely long drying times of one year or more were necessary to avoid the silica gels fracturing into a fine powder. As a consequence, there was little technological interest at this stage.

Initially there were a number of competing theories as to the structure and formation of silica gels, but by the early 1930's, the network structure was widely accepted. Hurd showed that they must consist of a polymeric skeleton of silicic acid enclosing a continuous liquid phase. In 1932, Kistler supercritically dried a silica gel and demonstrated the existence of the solid skeleton of the gel. Mineralogists at this time became interested in the use of sols and gels for the preparation of homogeneous powders for use in studies of phase equilibria, while sophisticated work was going on in the nuclear-fuel industry to try to prepare small spheres (10's of μm in diameter) of radioactive oxides that would be packed into fuel cells for nuclear reactors. In the late 60's and early 70's, the ceramics industry showed interest in gels. Controlled hydrolysis and condensation of alkoxides for the preparation of multicomponent glasses was discovered, and ceramic fibres were made from metal-organic precursors. However, it was in the mid-seventies that the breakthrough, which caused the explosion of activity that continues today, occurred.

There has been renewed interest on the part of researchers in the sol-gel process for making glass at lower temperatures.³ This is due to several factors: rising energy costs, recent advances that have made it more practical, and its potential

for producing the types of specialised glasses needed for new high-technology applications such as solar cells, fibre optics, lasers, and high-strength reinforcing fibres. Optical coatings that withstand high temperatures and rigorous conditions are very important for a number of present-day applications including optical sensing.

In this chapter, the sol-gel process will be explored. In principle, it is a relatively simple, low temperature method of materials fabrication. However, there are many parameters which can be varied resulting in materials with different properties. Each component / parameter will be examined and its effect on the process illustrated, thereby demonstrating the versatility of the process.

3.2 Outline of the process

The principle behind the sol-gel process for making glasses is one of building up reactive units in solution. These units form chains, rings and networks that are later present in the gel. But what exactly is a sol-gel material? A *sol* is a colloidal suspension of solid particles in a liquid, and is the first phase in the process. *Colloids* are solid particles with diameters of 1-100 nm. A *gel* is an interconnected, rigid network with pores of submicrometer dimensions and polymeric chains whose average length is greater than a micrometer. Silica gels can be fabricated in two ways: method 1 involves network growth from an array of discrete colloidal particles, and method 2 involves formation of an interconnected 3-D network by the simultaneous hydrolysis and polycondensation of an organometallic precursor. Method 2 is used in this work. A *precursor* is a starting compound which is composed of a metal or metalloid element surrounded by various ligands. The most common silicon precursor is tetraethoxysilane (TEOS), a *silicon alkoxide*, where the silicon atom is the central metal ion which is surrounded by four ethoxy groups. Another type of precursor which will be used in this work is an *organoalkoxysilane* compound, an example of which is methyltriethoxysilane (MTEOS). As before, this compound consists of a central metal ion (in this case silicon) which is again surrounded by ligands, but in this case one or more of the ligands is an organic group, hence they have direct metal-carbon bonds and not metal-oxygen-carbon bonds as in metal alkoxides.

A sol-gel material is fabricated from the combination of an inorganic or organic (or both) precursor, water, a catalyst, and a solvent such as ethanol. These

components are mixed well, whereby hydrolysis and polycondensation of the precursor occurs, resulting in the formation of a low density solid gel. This gel can then be subjected to a temperature programme which controls the densification process.⁴ A schematic of the process is shown in Figure 3.1.

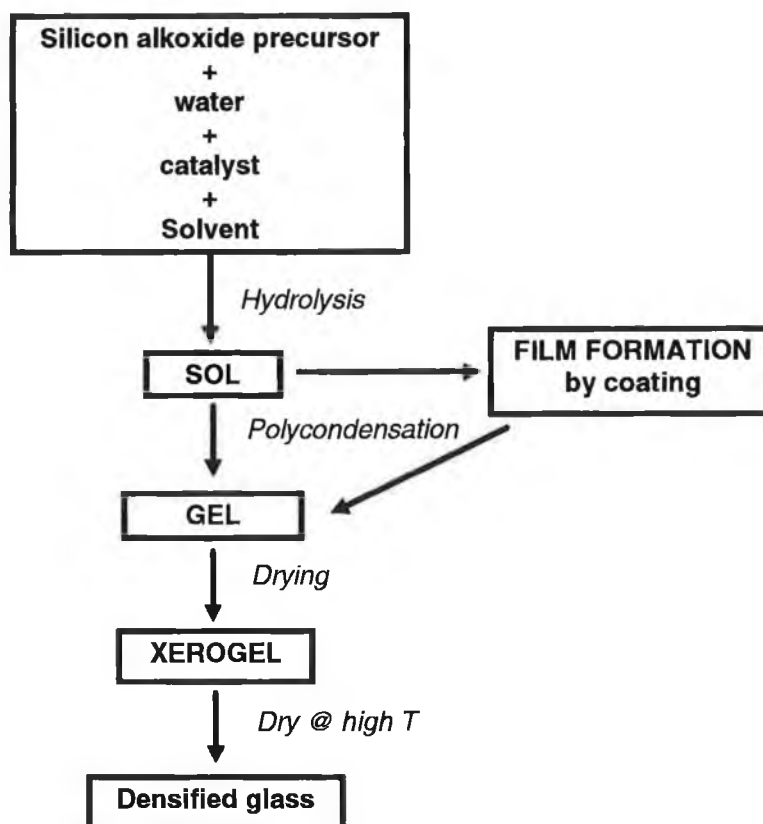


Figure 3.1: Outline of sol-gel process for the fabrication of glass.

The process is very well adapted for film formation as, at the sol stage thin glass films can be formed by dip-coating or spin-coating. These films, when dried at relatively low temperatures, are highly porous and are the basis of the sensors discussed in this work.

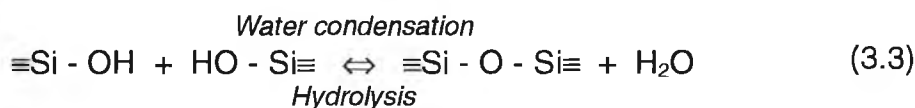
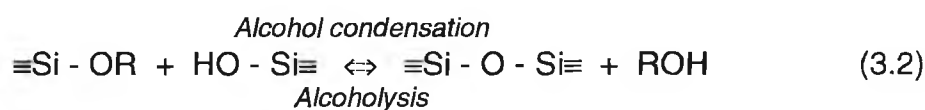
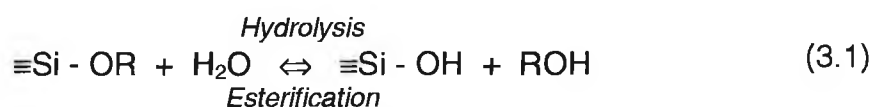
A *monolith* is a gel which has been cast into a mould and has its smallest dimension greater than a few millimetres. Hence a silica gel can be cast into fibres or moulded into lenses or other optical components.³ The sol-gel process can be used to make sols which are used to coat substrates such as protective coverings, anti-reflective coatings, selectively permeable membranes or it may be used as an adsorbent. The versatility of this process is endless. In the following sections, details of the process and the precursors will be outlined, and the effect of varying the process parameters with respect to each other will be explained. Firstly, the

basic hydrolysis and condensation processes will be described, and then it will be shown how the kinetics of these reactions are strongly influenced by the parameters, and that changing these will change the structure of the sol-gel end-product.

3.3 Hydrolysis and condensation polymerisation

A gel is synthesised by hydrolysis and polycondensation of organometallic compounds (e.g. alkoxides) which are dissolved in alcohols in the presence of a limited amount of water, using a mineral acid or base catalyst.

At the functional group level, three reactions are generally used to describe the sol-gel process:



where R is an alkyl group, $\text{C}_x\text{H}_{2x+1}$. The hydrolysis reaction (Eqn. 3.1) replaces alkoxide groups, OR, with hydroxyl groups, OH, and this occurs by the nucleophilic attack of oxygen contained in water on the silicon atom.² Subsequent condensation reactions involving the silanol groups occur via a nucleophilic condensation reaction and produce siloxane bonds, $\equiv\text{Si}-\text{O}-\text{Si}\equiv$, plus the bi-products alcohol, ROH, (Eqn. 3.2) or water (Eqn. 3.3). Water release is favoured unless water concentrations are very low, when alcohol liberation becomes significant. Under most conditions, condensation commences before hydrolysis is complete. Because water and alkoxysilanes are immiscible, a mutual solvent such as alcohol is normally used as a homogenising agent. However, alcohol is not simply a solvent, it can also participate in esterification or alcoholysis reactions as indicated in Eqns. 3.1 and 3.2. The reactions in Equations 3.2 and 3.3 proceed and build up long polymeric chains of $\equiv\text{Si}-\text{O}-\text{Si}\equiv$ molecules which interlink and form networks extending throughout the sol, leading to gel formation.

3.4 Factors affecting structure

There are a number of variables which affect the hydrolysis and condensation rates and hence the microstructure of the gel namely temperature, nature and concentration of the catalyst (acid or base), nature of the solvent, type of alkoxide precursor and ratio of water to precursor, concentration of the reactants, addition sequence, time of mixing, and pressure, although pressure is not usually a processing variable. All of these factors play an integral part in the formation of the microstructure of the sol-gel material by varying in different ways the hydrolysis and condensation reactions. The factors most relevant to this work are catalyst pH, and water to precursor ratio.

3.4.1 Influence of catalyst and pH

Hydrolysis and condensation are more rapid and complete when catalysts are employed. The pH-dependence of the gel-time, which is often used as a measure of the overall condensation kinetics for sol-gel systems [gel time $\propto 1/(\text{average condensation rate})$] can be seen in Figure 3.2.

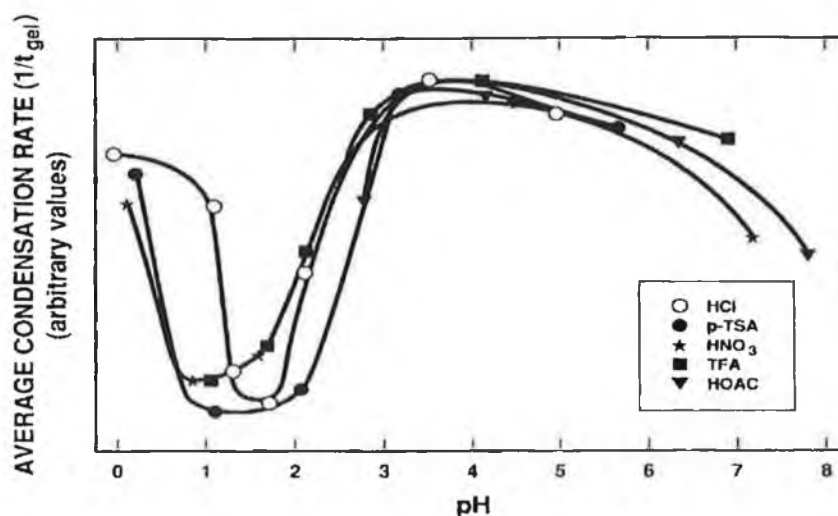


Figure 3.2: Average condensation rates ($1/t_{\text{gel}}$) for TEOS hydrolysed with solutions of various acids.²

According to this, the overall condensation rate is minimised at approximately pH 1.5-2.0 and maximised at intermediate pH.² The isoelectric point of silica, at which the electron mobility and the surface charge is zero, occurs at approximately pH=2. This pH value forms the boundary between so-called acid catalysis of the polymerisation process ($\text{pH} < 2$) and base catalysis ($\text{pH} > 2$). Under acid-

catalysed conditions, $\text{pH} < 2$, (fast hydrolysis and slow condensation), the gel formed consists of a fine network structure of linear chains, whereas under base-catalysed conditions (slow hydrolysis and fast condensation), more dense colloidal particles with large interstices between them are formed.⁵

Acid-catalysed reactions proceed according to Fig. 3.3.

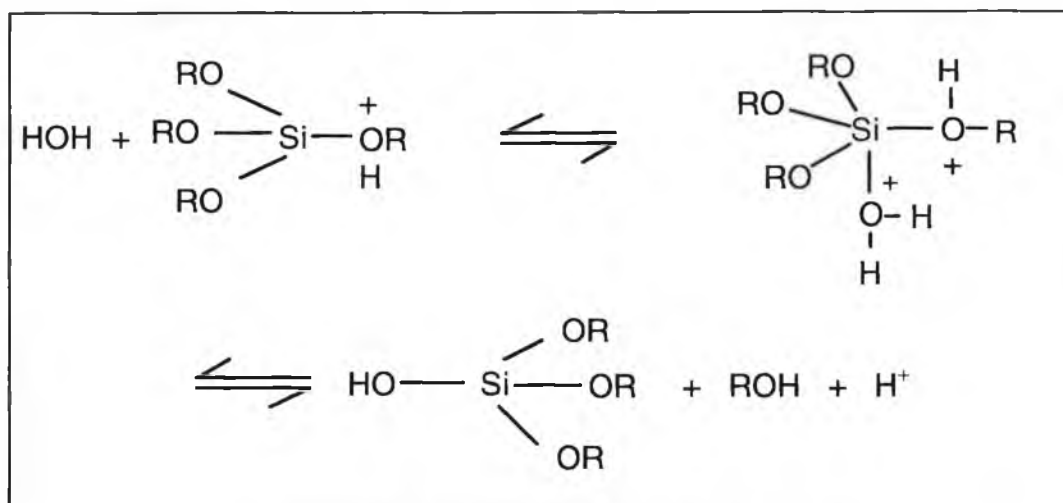


Figure 3.3: Acid-catalysed hydrolysis reaction.

In this reaction sequence, the alkoxide is protonated by the acid, thus increasing the acidity of the group and allowing the central silicon atom to be attacked from the rear by a water molecule. The water acquires a partial positive charge and consequently partially reduces the charge on the alkoxide, thus making it a better leaving group.⁶ Si-OH monomers cluster to form a weakly branched microporous structure with pore sizes $< 2 \text{ nm}$.

In the base-catalysed reaction, which may be seen in Fig. 3.4, the hydroxyl anion directly attacks the electropositive silicon, resulting in a partial negative charge developing on the silicon atom. Redistribution of charge occurs with the partial negative charge being accommodated by an alkoxy group. The alkoxy group is then able to leave the reactive intermediate, a proton being extracted from a readily available water molecule, with additional hydroxide ions being generated in the process.⁶

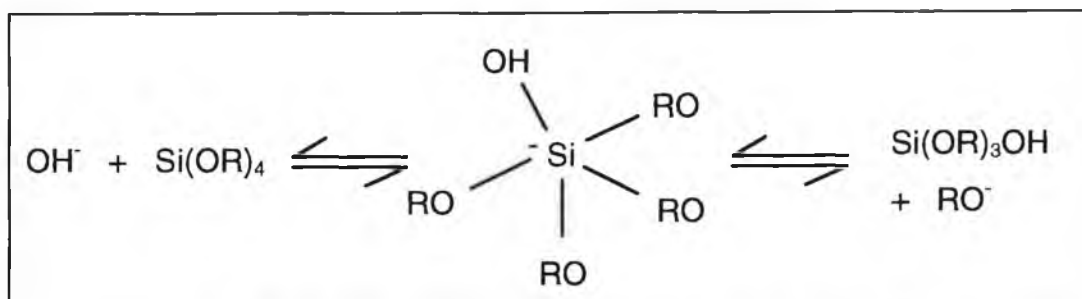


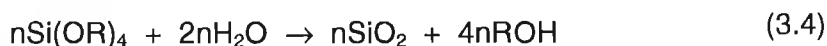
Figure 3.4: Base-catalysed hydrolysis reaction scheme.

The rate of base-catalysed hydrolysis is a function of the hydroxyl ion concentration in solution. Under basic conditions, hydrolysis is slow and condensation rates are faster, resulting in shorter gel times. The microstructure, in this case, consists of a highly-branched colloidal network with larger pores.

The pH of the system is a major factor in determining the porosity, hence when designing a system for the fabrication of a glass structure, pH is an important consideration. Acid-catalysed gels have greater flexibility to bend and rotate due to their network structure of long, thin chains, but are also subject to deformation. They form gels with small pores. On the other hand, base-catalysed gels have lower shrinkage, larger pores and higher porosity, and at the extreme conditions of very high pH, spherical or disc-shaped particles of uniform size are formed by what is commonly known as the Stöber process.⁷ Water to precursor ratio (R-value), as will be seen in the next section, also plays an important role in pore formation.

3.4.2 Influence of water:precursor (R) ratio

The R value is the molar ratio of water to silicon alkoxide precursor, $[R = (\text{H}_2\text{O})/(\text{precursor})]$. It plays a major role in the structural evolution of the sol-gel material. The size of the sol particles and the cross-linking within the particles (i.e. density) are primarily dependent on pH and on the R value.¹ Because water is produced as a by-product of the condensation reaction, an R value of 2 is theoretically sufficient for complete hydrolysis and condensation to yield anhydrous silica as shown by the net reaction:²



However, this stoichiometric value of 2 has been shown to be inadequate for the hydrolysis reaction to proceed to completion, due to reverse reactions and a number of competing processes. Hence, R values of greater than 2 are required

for hydrolysis to be completed before condensation and drying occur.⁴ From Equation 3.1, an increased value of R is expected to promote the hydrolysis reaction, which is the most obvious effect. Generally with under-stoichiometric additions of water ($R < 2$), the alcohol-producing condensation reaction (Eqn. 3.2) is favoured, whereas the water-forming condensation reaction (Eqn. 3.3) is favoured when $R \geq 2$.² Klein and Garvey found that for bulk gels, increased water increases the hydrolysis rate in both acid- and base-catalysed solutions. They also found that in acid-catalysed solutions, water has a strong effect on surface area and porosity, while acid addition has little effect.⁸

At pH 1, hydrolysis is relatively fast compared with condensation, and as R is increased, the extra water promotes hydrolysis and is consumed. At large R values and low pH, more efficient hydrolysis decreases the gel time resulting in thicker films. As R is increased further to the point where there is excess water over that required for hydrolysis at a given pH, the extra water serves to dilute the sol, reducing the relative silica content and giving rise to longer gel times and thinner films.⁹ Figure 3.5 illustrates this point.¹

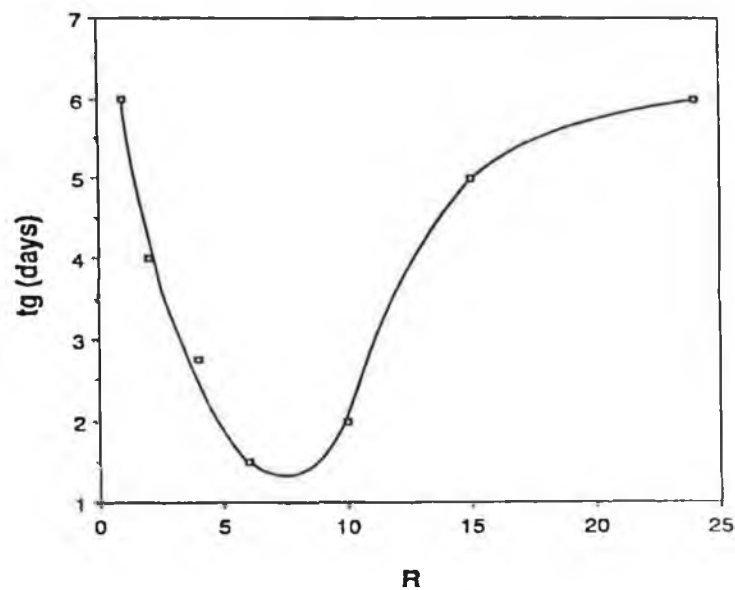


Figure 3.5: Variation of the gelation time with R ratio.¹

Experiments carried out by Colby et al¹⁰ show that for a R value of 2, the gelation time, t_g , is about 7 hours, and this decreases to 10 minutes for $R=8$. The location of the minimum in the curve is dependent on the experimental conditions, in particular pH of the reaction.

3.5 Film formation

The two most commonly used techniques of thin film production are dip-coating and spin-coating onto planar substrates. Dip-coating involves withdrawing a substrate vertically from a coating bath at a constant speed. The moving substrate entrains the liquid in a fluid mechanical boundary layer that splits into two above the liquid bath surface, returning the outer layer to the bath.¹¹ Since the solvent is evaporating and draining, the fluid film acquires an approximate wedge-like shape that terminates in a well-defined drying line. When the receding drying line velocity equals the withdrawal speed, the process is in steady state with respect to the liquid bath surface. Figure 3.6 illustrates the various stages in the dip-coating process.

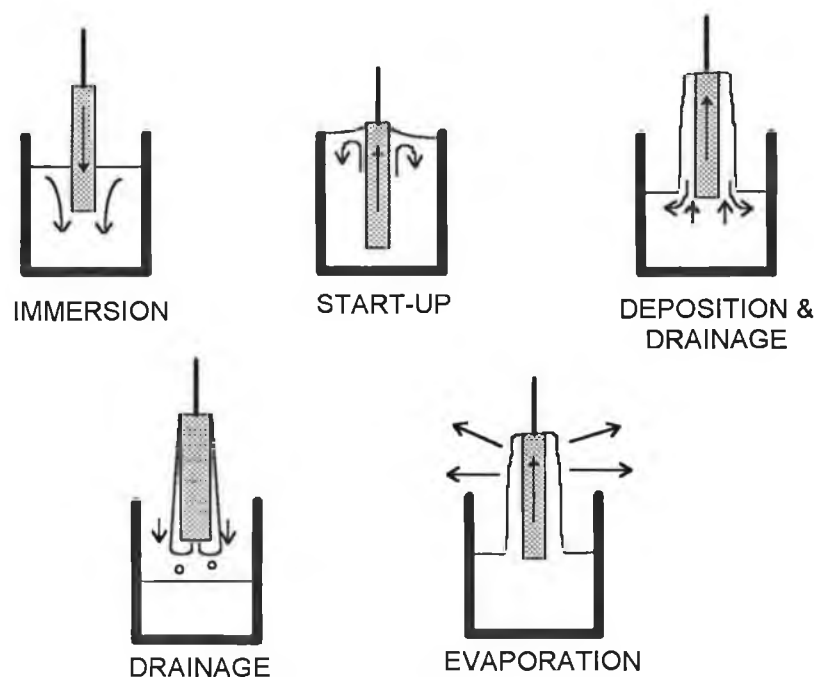


Figure 3.6: Stages in the dip-coating process.²

For alcohol-rich fluids common to sol-gel dip-coating, steady state conditions are attained in several seconds. A competition between as many as six forces in the film deposition region governs the film thickness and the position of the streamline: (1.) viscous drag upward on the liquid by the moving substrate; (2.) force of gravity; (3.) resultant force of surface tension in the concavely-shaped meniscus; (4.) inertial force of the boundary layer liquid arriving at the deposition region; (5.) surface tension gradient; (6.) the disjoining (or conjoining) pressure.

The following equation denotes the relationship between film thickness, t , and dip speed, U , for Newtonian fluids¹²

$$t = 0.944 (N_{ca})^{1/6} \left(\frac{\eta U}{\rho g} \right)^{1/2} \quad (3.5)$$

where ρ is the solution density, g is the acceleration due to gravity, η is the dynamic viscosity and N_{ca} is the capillary number, defined as $N_{ca} = (\eta U)/\sigma$, where σ is the surface tension. Clearly if the viscosity and density remain constant, film thickness is proportional to withdrawal speed therefore thickness increases with dip speed.⁹

Spin-coating involves the deposition of thin films by centrifugal draining and evaporation. An advantage of spin-coating is that a film of liquid tends to become uniform in thickness during spin-off and once uniform, tends to remain so, provided that the viscosity is not shear-dependent, and does not vary over the substrate. Also due to the small amount of liquid required for spin-coating, there is little waste. However, this method does not work well for very large substrates or non-circular substrates, whereas dip coating works well for any shape or size of substrate.

The main differences between dip- and spin-coating is the evaporation rate and the time available for aging or prepolymerisation (see Section 3.5.1). Spinning creates a strong forced convection in the vapour above the substrate increasing the evaporation rate. Thus there is little time for aging to occur, and the structure of the film is dominated by the effects of capillarity.¹¹

According to Eqn. 3.5, the thickness of the film is altered by varying the dip speed, hence the film structure may be controlled by dip speed for a particular sol.^{11, 13} Strawbridge et al¹² showed that when $\log(\text{film thickness})$ is plotted against $\log(\text{dip speed})$, a slope of 0.5 or 0.66 depending on assumptions related to surface tension and viscosity dependence is obtained in agreement with Eqn. 3.5. Films whose thickness exceed 600 nm are poor quality and non-uniform. This is due to drying stresses within the microstructure. Drying films at very high temperatures can stress the film due to the quick evaporation of water and/or solvent from the pores which leads to cracking.

3.5.1 Film aging and prepolymerisation

Aging is the process that takes place after mixing the precursor, water, solvent and catalyst to form a sol, but before dip-coating, in the case of coating sols. The sol

is left at room temperature (in ambient conditions) or at elevated temperatures in order to accelerate the hydrolysis and condensation process. This aging process serves a number of purposes. Aging for a monolithic sol is different to that of a coating sol. A monolithic sol can be aged before or after gelation. Before gelation, polymerisation takes place and the viscosity increases. After gelation, polycondensation continues along with localised solution and reprecipitation of the gel network which increases the thickness of interparticle necks and decreases porosity.⁶ The reason for doing this is to produce a gel of sufficient strength to withstand the stresses encountered during drying. For coating sols, the main function of aging is to increase the viscosity of the solution by accelerating cross-linking of polymers to ensure that dip-coating is possible. If a sol is not viscous enough, it will not coat a planar substrate. Depending on the duration of aging, various film thicknesses can be achieved as a result of varying the sol viscosity. Aging has also been found to promote film porosity.²

3.5.2 Film drying and gelation

Hydrolysis and condensation reactions lead to the growth of clusters that eventually collide and link together into a gel. The sol becomes a gel when it can support a stress elastically. This is typically defined as the gelation point or gelation time, t_g .¹ For thin films, gelation begins soon after coating. The chemical reactions that bring about gelation continue long beyond the gel point, permitting flow and producing gradual changes in the structure and properties of the gel.

After dip-coating, films can be dried at room temperature or at elevated temperatures. Even at room temperature, the drying process for films is much more rapid than for monolithic sol-gel materials, due to the relatively small volume of sol in the film. In general, films are more dense and less porous than monolithic materials. Porosity and density can, however, be controlled to a certain extent by varying processing parameters as mentioned previously, as well as by varying the drying time and temperature. In this study, the required film porosity was achieved by drying the films at 70°C for 18 h, as discussed in Chapter 4.

3.6 Organically modified precursors

The surface of a standard alkoxide-based gel or sol-gel film is largely covered with hydroxyl groups. These hydroxyl groups are the negative ion part of the surface silanol (SiOH) groups formed by hydrolysis of the silicon alkoxide precursor, as seen in Eqn. 3.1. As a consequence of the silanol groups, the surface is hydrophilic and allows the adsorption of a layer of water molecules, by physisorption, which coats the inside of each pore. Some applications require the surface to be hydrophobic which necessitates the removal of the hydroxyl groups. Water is present in two forms on the film. The first is 'free' water within the ultraporous gel structure, physisorbed water, and the second is hydroxyl groups associated with the gel surface i.e. chemisorbed water. The amount of physisorbed water adsorbed onto the silica particles is directly related to the number of hydroxyl groups existing on the surface of the silica.¹ By removing the surface hydroxyls, not only is the chemisorbed water removed but also the physisorbed water, which does not exist on the surface without the presence of hydroxyl groups.

There are number of ways of removing surface hydroxyls, of which the most important are thermal dehydration and chemical modification. For the fabrication of porous films, thermal dehydration is unsuitable due to the structural densification and collapse of pores. Chemical modification is a much more suitable method for this application, as it removes the hydroxyl groups making the surface essentially hydrophobic, while retaining the porosity.

Chemical modification involves the combination of organic and inorganic precursor materials on a nanometre scale. The materials formed are intermediate between glasses and polymers.¹⁴ The modified materials are known as *ORMOCERS* (*Organically Modified Ceramics*) or *ORMOSILS* (*Organically Modified Silicates*).¹⁵

Organically substituted alkoxysilanes, such as $R'Si(OR)_3$, where R' is the alkyl substituted group and OR is the standard alkoxy group, form inorganic/organic networks by hydrolysis and condensation reactions. Instead of forming surface silanol (Si-OH) groups, the alkyl group takes the place of the hydroxyl and forms Si-R groups, which are neutral, giving rise to a hydrophobic surface with increased contact angle. Kron et al¹⁶ describe how the contact angle can be increased by increasing the amount of modified precursor in the sol. It has been shown using Fourier-Transform Infrared (FTIR) Spectroscopy that the addition of organically modified precursors increases the Si-R groups while decreasing the Si-OH band at

3740 cm^{-1} and molecular water band at 3400 cm^{-1} .^{17, 18} This will be described in more detail in Chapter 6.

In this application, organically modified precursors were used to fabricate films for use as dissolved oxygen sensors. As will be seen in later chapters, hydrophilic TEOS-based films did not respond well in water due to adsorbed water on the pore surface. Organically modified films, however, due to their hydrophobicity excluded the water which, in turn, improves their response to oxygen in water.

3.7 Encapsulation of organic molecules

Dye molecules have been used to sense various analytes by many groups. Porous Vycor glass or RTV silicone rubber have been post-doped with organic dye complexes. They have been covalently bound to polymers and have also been embedded in polymeric substrates. Fibres have been etched and organic dyes adsorbed onto the pore surface.

Sol-gel glass is a very suitable host matrix for dye complexes. Not only can porous glasses be post-doped / impregnated with an organic dye, but they can more importantly be encapsulated during fabrication without degradation or damage to the complex. Post-doping is highly versatile, but the attachment of the reagent to the support is weak which limits the practical operation and shelf-life of these sensors.¹⁹ Impregnated devices are useful for continuous operation only when the driving force for leaching is low, such as when water is excluded by a hydrophobic matrix, and for gas sensing. Encapsulation of the dye, however, means that the dye is entrapped in the porous silica structure. It is thought that the dye complex is almost fully accessible to the analyte species via the pores, as will be seen in later chapters. The exact location of the dye complex in the sol-gel structure is not known, but due to the absence of leaching occurring, it is thought that the structure forms around the complex. Sol-gel glasses are chemically durable and do not degrade, and in the case of the ruthenium complex used here, leaching of the chromophore or photodegradation do not occur.

As mentioned in Chapter 2, the chromophore used in this work is a transition metal compound, ruthenium-tris(4,7-diphenyl-1,10-phenanthroline)dichloride, which is an oxygen-sensitive complex. For the fabrication of oxygen-sensitive films, the dye complex is dissolved in ethanol, after which water is added and mixed using a magnetic stirrer. The final component to be added is the silicon alkoxide

precursor, which is added dropwise while stirring. In this way, the dye complex is encapsulated within the matrix. The concentration of dye used is a 40,000 ppm of dye by weight per silicon atom, which is 2.5 g/L of solution. This is quite a high concentration of dye, and exhibits strong fluorescence under blue LED excitation.

3.8 Summary

The sol-gel process has been described in detail in this chapter, both the chemical processes of hydrolysis and polycondensation, and the variety of fabrication parameters which affect these reactions and determine the final structure of the materials. A description of how the films can be chemically modified to remove the surface hydroxyl groups, and render the surface hydrophobic was described. The encapsulation of the oxygen-sensitive dye complex has been detailed, and the position of these molecules in the structure discussed. The advantages of encapsulation over impregnation have also been discussed, particularly for this application.

The next chapter details the preparation and fabrication of these sensing elements. The substrate preparation and cleaning, both for planar and fibre substrates is described, after which sol fabrication and film formation are discussed.

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CHAPTER 4: SENSOR FABRICATION

4.1 Introduction

Chapters 2 and 3 have dealt with the oxygen-sensitive ruthenium metal complex and its properties, and the sol-gel fabrication process, respectively. In this chapter, the two are combined and the specific film fabrication process for this application is detailed.

DO sensors must be sensitive to low oxygen concentrations. The response must be repeatable and fully reversible i.e. the quenching response must remain constant each time the sensor is used, and the fluorescence intensity in the absence of oxygen must be the same after numerous cyclings through oxygenated and deoxygenated water. Hence, it is necessary that photobleaching and leaching do not occur. The sensor must be physically rigid and must not degrade in water. It is important that the fabrication process is simple and reproducible to enable reproduction of numerous batches of sensing films with identical properties.

In this chapter, the procedure for obtaining coating sols is described, along with the method of preparation of coating substrates. Coating is performed using a dip-coating apparatus, which is illustrated, and the films are optimised with respect to dye concentration.

4.2 Preparation of a ruthenium-doped sol

As outlined in Chapter 3, the preparation of a doped sol involves the mixing of a silicon alkoxide precursor, alcohol, water, an acid catalyst and the dye complex in the correct proportions to form a coating sol. The preparation of a TEOS-based sol is detailed below. It is an acid-catalysed sol with R value of 2, consisting of a network of linear polymers, which when dip-coated and dried forms a microporous film. The method of preparation is as follows: 0.0323 g of $[\text{Ru}^{(\text{II})}(\text{Ph}_2\text{phen})_3]\text{Cl}_2$ is placed in a clean vial. Into this is weighed 4.1508 g of ethanol, and the contents are stirred well using a magnetic stirrer. When the dye complex has dissolved in ethanol, 1.0376 g of water with a pH value of 1 is added to the vial, and this is stirred again. Finally, 6.0000 g of tetraethoxysilane (TEOS) is added dropwise while stirring to ensure a homogeneous solution. The mixture is then left to stir for

1 h, before removing the magnetic bob and aging at 70°C for 18 h. During film aging, the vial is sealed to prevent fast evaporation and subsequent gelation, but a tiny hole in the lid of the vial allows for slow evaporation and pressure release. After curing, the sol is sufficiently viscous to dip-coat planar / fibre substrates, the withdrawal speed determining the film thickness. As was seen in Chapter 3, a faster withdrawal speed gives a thicker coating (Eqn. 3.5). The dip-coating apparatus will be shown in greater detail later in this chapter. Having coated the substrates, they are cured at 70°C for 18 h, to complete the polymerisation process and drive off excess water. The films are then stored in air-tight containers away from light before use.

The concentration of ruthenium complex in the sol is expressed in two ways in this work; as amount of ruthenium complex by weight per silicon atom, and as amount of ruthenium per total volume of solution. The normal concentration of ruthenium used in this work is 40,000 ppm or 2.537 g/L, as it gives strong fluorescence under blue excitation. Optimisation of this concentration will be discussed in Section 4.5. Films have been fabricated using different precursors and different R values. R=3 TEOS and R=4 TEOS films, were fabricated as well as modified films with increasing ratios of modified precursor to TEOS, ranging from films with 100% TEOS to films with 100% modified precursor. The modified precursors used in this work were TEOS-based, having silicon as the central metal ion with three ethoxy (-OC₂H₅) groups. The fourth ethoxy group is substituted for an organic group i.e. a ≡Si-C- bond is formed instead of a ≡Si-OC- bond. These precursors are as follows: triethoxysilane (HTEOS), which has a proton, H⁺, substituted for one of the ethoxy groups, methyltriethoxysilane (MTEOS), whose substituted group is a methyl (-CH₃) group, ethyltriethoxysilane (ETEOS) and phenyltriethoxysilane (PhTEOS) whose substituted groups are ethyl (-C₂H₅) and phenyl (-C₆H₅), respectively. For R value variation, the amount of precursor is varied keeping the water:ethanol ratio constant. Likewise for the variation of modified precursor to TEOS, basic calculations are used to determine the amounts of each constituent required.

4.3 Preparation of planar/fibre substrates for sensing

Both planar and fibre substrates were used in this work, although most of the work was performed using planar substrates. These planar substrates were of two kinds, soda-lime glass and silicon wafers. The glass slides were used for fluorescence quenching and leaching experiments and the silicon wafers were used for ellipsometric thickness measurements. The function of the coated fibres was to perform quenching experiments to demonstrate the viability of a fibre-based DO sensor.

The soda-lime glass substrates are cut to a size of approximately 35 mm x 15 mm from standard microscope slides, which are approximately 1 mm thick. A standard glass cutter is used for this procedure. For the silicon slides, however, a silicon scribe is used to cut the wafers. These substrates are slightly smaller than their glass equivalents due mainly to the relative scarcity of the silicon wafers and also as a large area film is unnecessary for thickness measurements. More recently, a microlitre cell was designed to perform response time experiments. The glass slides, specially manufactured along with the microlitre cell (Optiglass, Essex, U.K.), are larger than the previous substrates and are (40 x 14 x 1.5) mm. Before dip-coating, it is important to ensure that the substrates are clean and free from any oil or organics. In the literature there are many different cleaning procedures used, among which include cleaning with detergents and surfactants using rotating brushes and sonication.¹ The procedure used here is not as rigorous, however, but is adequate for this application.

Having cut the microscope slides to size, they are first washed with deionised water, then sequentially washed with methanol, acetone and finally deionised water again. The clean slides are then conditioned in deionised water at 70°C for 24 h, as this is thought to increase the number of surface silanol groups which should enhance adhesion of the sol-gel film to the substrate. This cleaning procedure applies to both glass and silicon substrates. The fibres, however, are treated differently.

The fibre used in this work is a plastic clad silica (PCS) fibre, of core/cladding diameter 600/760 μm and a numerical aperture of 0.4 (Ceramoptec Inc., Enfield, CT., U.S.A.). It is a large silicone-clad step-index fibre which is durable and easy to handle. It is relatively easy to remove the cladding, and also relatively easy to couple the light into the fibre from a laser diode or LED due to the large diameter. However, because the fibre diameter is so large, standard cleaving tools are

unavailable, hence the fibre must be cut into lengths using a fibre cutter to ensure minimum damage, and then polished at each end. The average fibre length was about 13 cm. A Logitech PM2A polishing rig was used to polish the fibre, and it polishes approximately 120 fibres at once.² It is imperative that the fibre ends be parallel to the polishing wheel. There are three steps in the polishing process. The first involves polishing with a steel polishing wheel while using $9\ \mu\text{m}\ \text{Al}_2\text{O}_3$ fluid. The grinding action of the fluid with the rotation of the steel polishing plate for about 3 hours polishes the end faces of the fibres. The second step involves the use of the steel plate with a $3\ \mu\text{m}\ \text{Al}_2\text{O}_3$ solution. Finally, the steel plate is replaced with a polyurethane wheel which is used with a $0.125\ \mu\text{m}$ polishing suspension for about 3 h. This is then repeated for the other end face, after which the cleaning and cladding removal takes place.

Each fibre is cleaned, first with water and then with alcohol to remove any Al_2O_3 . The primary coating is then carefully removed from approximately 8 cm of the fibre length using a scalpel. The silicone cladding is removed by etching using a commercially available solvent mixture of methylene chloride and sulphuric acid (Lumer, Bagnolet, France). The fibres are left in this etchant for about 15 minutes, after which they are removed and washed sequentially in water, methanol, acetone and deionised water. After cleaning, they are conditioned in the same fashion as for the planar substrates before dip-coating. A diagram of the structure of the fibre sensing substrate can be seen in Figure 4.1.

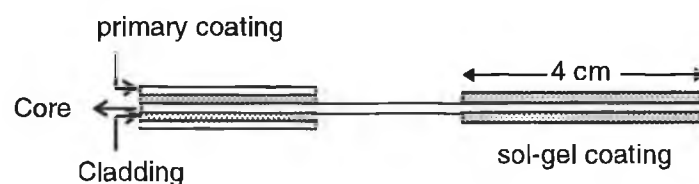


Figure 4.1: Structure of the sensing substrate

4.4 Dip-coating of sensing substrates

The theory behind the dip-coating process was detailed in Section 3.4.4. Here the basic apparatus and experimental procedures are described. Dip-coating is the most suitable method of coating substrates for this application as small planar substrates as well as fibre substrates can be used.

The dipping apparatus seen in Fig. 4.2 consists of a large frame on which the

substrate is held rigidly, and a moveable platform connected to a D.C. motor via a threaded bar. The motor controls the threaded bar, and as the bar turns the platform moves, hence the withdrawal speed can be controlled by varying the speed of the motor via the BBC computer. The vial containing the coating sol is held on the moveable platform such that the stage can be moved upwards to immerse the slide, and withdrawn at a constant rate depositing a film of thickness proportional to the withdrawal speed.

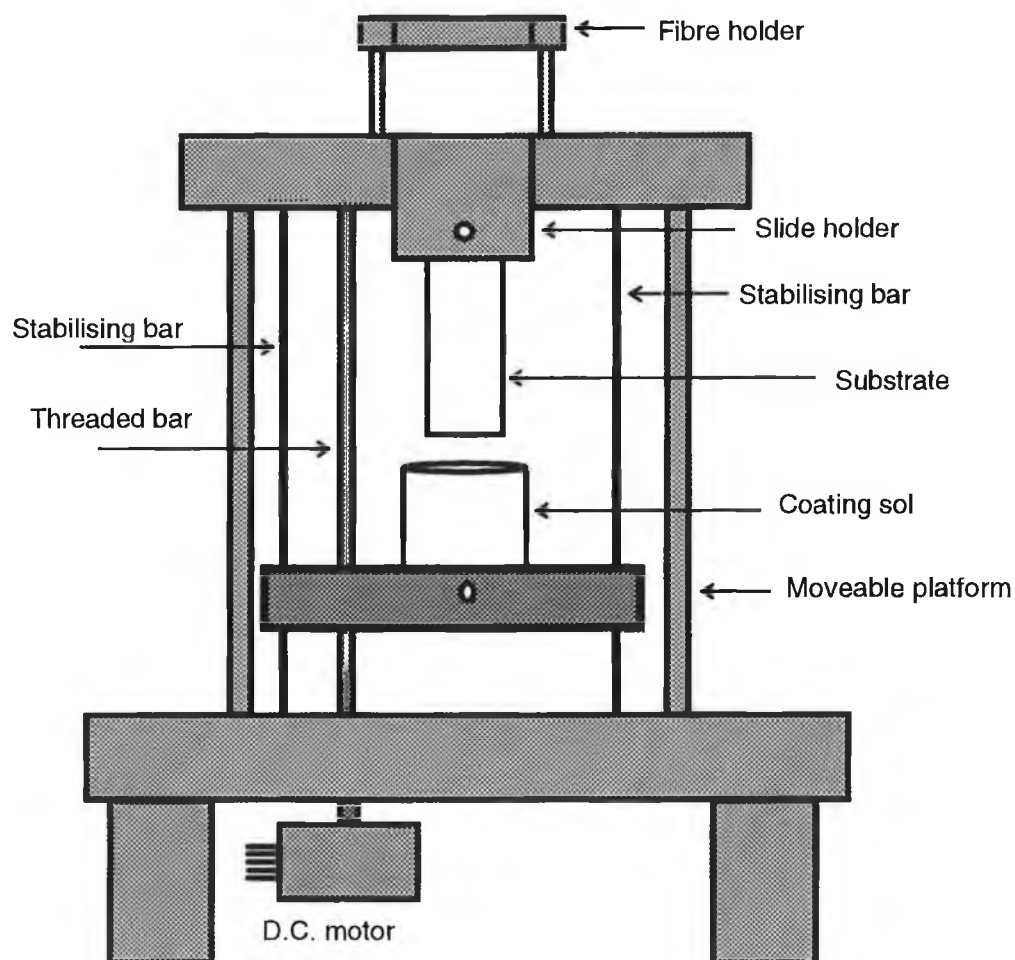


Figure 4.2: Apparatus for dip-coating planar and fibre substrates.

It must be noted that a good quality motor with absolutely smooth movement should be used in order to obtain high quality films. This apparatus is housed in a convection-free chamber closed to the atmosphere, and on vibration damping mats. The chamber's function is to avoid draughts which result in uneven films, and the damping mats ensure that the liquid surface remains completely stable

during the dip-coating process. After dipping, the coated substrates are placed on drying racks and held vertically while cured at 70°C for 18 h, during which polymerisation and solvent evaporation occur causing the structure to become rigid. The normal coating speed used is 1 mm/s, which results in a film 300-400 nm thick.

When dip-coating fibres, a portion at the distal end of the fibre, i.e. the end furthest from the excitation source, was not coated, as it was required to hold the fibre at both ends in the cell for stability. Prior to coating, a warm solution of paraffin wax in toluene was used to protect the distal end of the fibre. When this dried, the fibres were dip-coated, after which the sol-gel-coated paraffin wax was removed by dipping in a warm solution of toluene. This prevented a portion of sol-gel film which was inaccessible to the DO solutions from fluorescing and causing a large background.

4.5 Optimisation of dye concentration

There are a number of physical parameters regarding the sensing film which need to be optimised in order to design an optimal optical sol-gel-based sensor. Chapter 6 outlines film optimisation for DO sensing. It maps the progression of the sol-gel film from being a good oxygen gas sensor, through tailoring the microstructure to optimise a particular oxygen concentration range of interest, to surface modification and the achievement of a high sensitivity dissolved oxygen sensor. In this section, however, the optimisation of the dye concentration in the film is described.

The concentration of fluorophore in the sol-gel film for fluorescence-based dissolved oxygen sensing is optimised by investigating two quantities. The first is the absolute fluorescence intensity, which would be expected to increase with the concentration of dye complex, assuming dye leaching does not occur, until the onset of concentration quenching (Section 2.3.2). Monitoring the quenching response of the film in DO is the second method.

The ruthenium dye complex concentration was varied within the sol-gel films from 5,000 ppm (weight of ruthenium complex relative to the amount of silica), up to 100,000 ppm. 5,000 ppm was chosen as a minimum concentration, as fluorescence signals from films with less than this concentration were extremely low. Planar substrates were coated with the various sols and subsequently dried.

The absolute fluorescence intensity of the slides in fully deoxygenated water was measured using the system which will be seen in Section 5.3.1, after which their quenching response, Q_{DO} , was measured. Q_{DO} , as will be seen in Chapter 6, is a function of the fluorescence intensity in deoxygenated water and that in oxygenated water.

Figure 4.3 shows the absolute fluorescence intensity in deoxygenated water as a function of ruthenium complex concentration in the sol-gel film.

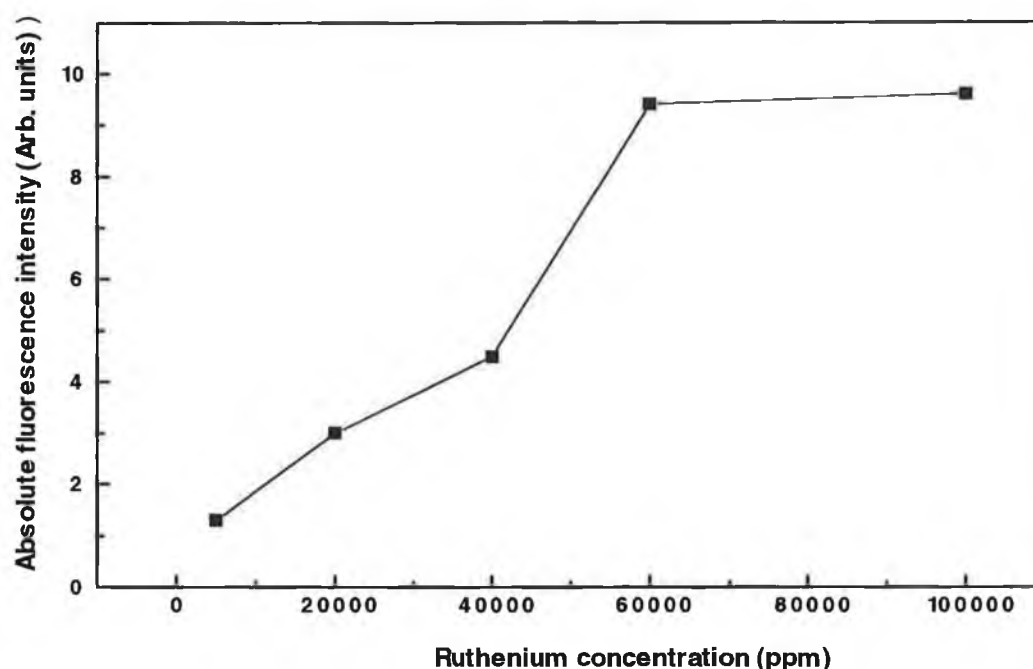


Figure 4.3: Absolute fluorescence intensity in deoxygenated water as a function of ruthenium complex concentration in the sol-gel film.

The absolute fluorescence intensity was measured in deoxygenated water to ensure that no quenching by oxygen was occurring. It can be seen in Fig. 4.3 that as the ruthenium complex concentration increases, so too does the absolute fluorescence intensity. Up to 60,000 ppm there is a large intensity increase, after which it levels off with only a small intensity increase from 60,000 ppm to 100,000 ppm, perhaps indicating a saturation level at approximately 60,000 ppm which may indicate the onset of concentration quenching. At 5,000 ppm there is a very low signal to noise ratio which steadily increases with oxygen concentration. The quenching response in water was also monitored for the same planar substrates, and the results are shown in Fig. 4.4. There is very little variation in quenching

response, of the order of 4 percentage units, from very low to very high dye concentrations. The quenching response increases and then decreases with increasing dye concentration. At 5,000 ppm dye concentration, the quenching response, Q_{DO} , was 70.5%, at 100,000 ppm, Q_{DO} was 70%, and it peaked at 74% for a dye concentration of 40,000 ppm. There was only a 1% difference between the peak value, and that of 20,000 ppm and 60,000 ppm, which is within the error for these measurements ($\pm 2\%$), where 2% represents the standard deviation of 5 readings.

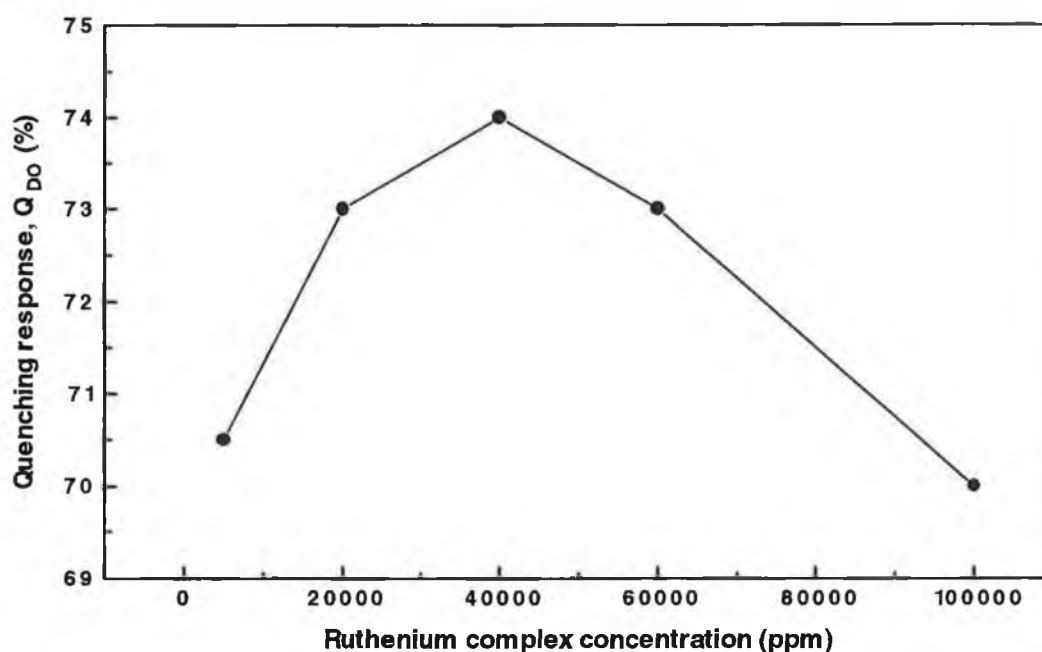


Figure 4.4: Quenching response in water as a function of ruthenium complex concentration.

The slight decrease in Q_{DO} at approximately 40,000 ppm is likely to be due to the decrease in accessibility to oxygen of individual ruthenium complex molecules as the concentration increases. It is likely that aggregation of the dye occurs above this concentration, leading ultimately to concentration quenching at higher concentrations as seen in Fig. 4.4.

By combining the results from Fig. 4.3 and 4.4, a dye concentration of 40,000 ppm was chosen as the optimum. The fluorescence intensity is sufficiently high under blue LED excitation for this dye concentration, resulting in a high signal to noise ratio. A further increase in the concentration of dye in the sol-gel film would result in a decrease in quenching response. As a result, for the work presented in this

thesis, a concentration of 40,000 ppm weight of ruthenium complex on silica, was used as the dopant concentration in the fabrication of dissolved oxygen sensors.

4.6 Conclusion

This chapter detailed the fabrication of both planar and fibre sensors for fluorescence-based dissolved oxygen sensing. Fabrication of the sensor involves preparing the sensing substrates by cutting, polishing and decladding in the case of fibres, and cleaning and conditioning the substrates so that they are ready for coating as was seen in Section 4.3. The method of preparation of coating films was outlined, and the apparatus used to dip-coat the films was illustrated and described in detail. The optimum ruthenium dye concentration in the sol-gel film was identified, and the optimisation experiments and data were shown.

Further optimisation data are shown in Chapter 6, which involve variation of the silicon alkoxide precursor or the organically modified precursor to optimise the films both for a particular application and within a particular concentration range of interest.

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CHAPTER 5: EXPERIMENTAL CHARACTERISATION SYSTEMS

5.1 Introduction

The sensor under development in this work is one based on fluorescence i.e. the sensor responds either to changes in fluorescence intensity or lifetime, or to the phase shift in fluorescence. In this chapter the various systems used to monitor the sensor as a function of fluorescence intensity, fluorescence lifetime or phase angle are described and illustrated. Figure 5.1 shows a basic system for measuring fluorescence intensities. The basic components include an excitation source, focusing optics, fluorescent material, collection optics, filters and a detector. The emission spectrum of the source should overlap the excitation band of the material, λ_{ex} , such that it excites the material causing it to fluoresce. A broadband source may be used, e.g. a white light source, but it will be necessary to use a filter or filters to select the suitable band in order that efficient excitation occurs without contributions from scatter or stray light. The excitation light is focused onto the sample using focusing lenses, resulting in fluorescence. Collection optics are used to capture the fluorescence, λ_{em} , which is filtered again to minimise detection of excitation light, with minimal reduction of the fluorescence signal. The fluorescence is then focused onto a photodetector which along with dedicated electronics records the detected light as a function of time.

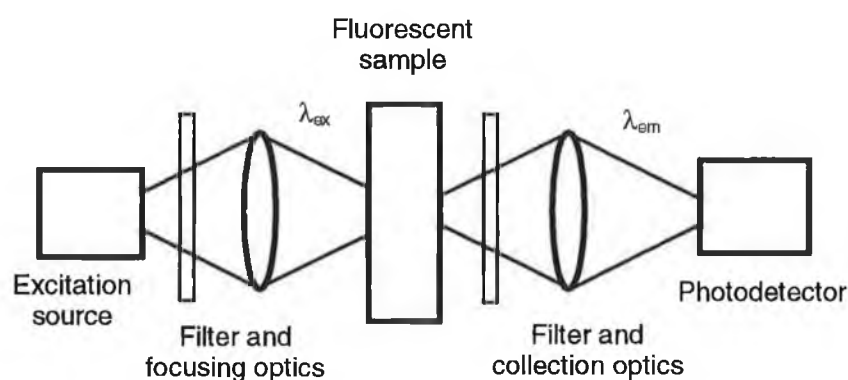


Figure 5.1: A typical system for measuring fluorescence intensity

This system can be used to monitor fluorescence lifetimes by substituting the continuous excitation source for a pulsed source, and ensuring that the detector is fast enough to measure the decay. Phase fluorimetric measurements can be made by modulating the excitation source and using a phase-sensitive detector.

Preliminary measurements were performed in gas phase to investigate the viability of the technique for sensing oxygen. These experiments were carried out using a laser-based system, as were the preliminary dissolved oxygen experiments. Due to the increasing availability at that time of blue light emitting diodes (LEDs), it was feasible to miniaturise the system by substituting a blue LED for the laser, and incorporating optical filters in place of the spectrometer.

This chapter traces the development of the system from a laser-based gas/aqueous phase system to a miniature portable system demonstrating the feasibility and relative simplicity of fluorescence intensity and phase fluorimetric measurements in either planar or fibre configurations.

5.2 Laser-based systems

5.2.1 Fluorescence intensity system

Preliminary gas and aqueous-phase measurements were made using a laser as excitation source. This apparatus can be seen in Fig. 5.2. Gas-phase measurements were the first investigative experiments to be carried out on sol-gel sensing films. An air-cooled argon-ion (Ar^+) laser (Omnichrome, MWK Industries, Corona, CA.) was used as the excitation source in this system, and light containing all the lines in the Ar^+ spectrum passes through a 488 nm line filter, A, (Optometrics U.K. Ltd., Leeds, U.K.).

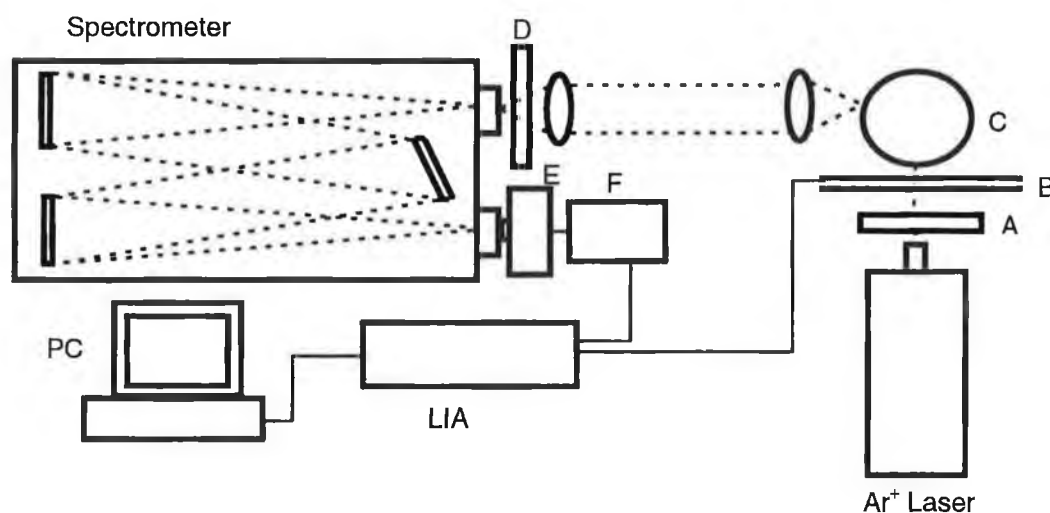


Figure 5.2: Laser-based fluorescence intensity system.

The excitation light excites the sol-gel-coated glass substrate at an angle of 45 degrees in the gas cell, C. The chopper, B, modulates the light at a particular frequency, providing a reference for the lock-in amplifier which prevents interference by ambient light. The fluorescence is collected and focused onto the entrance slit of the spectrometer, having passed through a long-wave pass glass filter, D, ($\lambda_{\text{cut-on}} = 560 \text{ nm}$ and having a transmittance of 83.9% at 608 nm). The fluorescence passes through a 1 metre Jobin-Yvon spectrometer (Model THR 1000) in a Czerny-Turner configuration, whose wavelength is set at 608 nm, the peak of the fluorescence spectrum. Essentially the spectrometer is acting as a filter. At the exit slit, a photomultiplier tube, E, (Hamamatsu, Middlesex, U.K.; Model R928) is housed, which detects the fluorescence. The signal is amplified using a pre-amplifier, F, after which it is fed into a lock-in amplifier (EG&G, Princeton, NJ., U.S.A.; Model 5206), along with the reference signal from the chopper, B, for synchronous detection. The analog signal is converted to digital by passing to a Bytronics multifunction input / output card (Model MP1BM3) and the digital data is then saved in a data file in the computer.

The gas cell, C, containing the sol-gel-coated substrate, is connected to a gas mixing system, which can be seen in Figure 5.3.

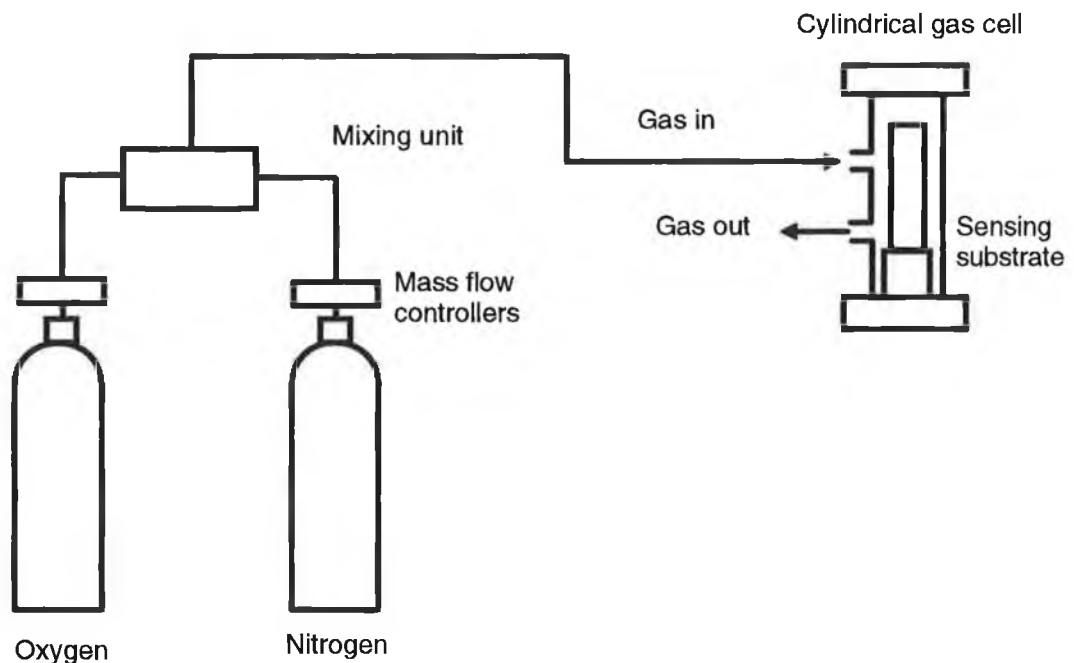


Figure 5.3: Gas mixing system with gas characterisation cell for laser-based experiments.

The gas system consists of two gases, oxygen and nitrogen, supplied from pressurised gas cylinders. These gases flow through mass flow controllers (Unit Instruments, Dublin, Ireland; Model UFC-1100A), which have been calibrated for each of the respective gases for a flow range of 0 to 500 sccm, where sccm denotes the units of rate of flow, standard cubic centimetres per minute. The flow of the gases and the mixing ratio is determined by a two-channel power supply / controller unit (Unit Instruments; Model URS-40), whereby the flow can be varied linearly between 0 and 100% flow for each gas. Oxygen and nitrogen are mixed using the controller unit to obtain varying concentrations of oxygen for measurement, while ensuring that the flow rate is kept constant. The exhaust from the mass flow controller passes through a tube and into the gas cell for measurement. As the concentration of oxygen increases, the fluorescence intensity decreases. This is a non-linear dependence and is characteristic of the quenching process as can be seen from the Stern-Volmer equation (Eqn. 2.12). Preliminary dissolved oxygen measurements were performed using the same laser-based characterisation system. The measurement cell, however, was very crude, consisting of a small volume glass vial with a plastic lid containing water. The oxygenated and deoxygenated water was created by bubbling the gas into the water in the vial. Due to the low volume of water, equilibration takes only a short time. The sensing substrate was mounted in the plastic lid, through which the gas inlet tube and a hypodermic needle for pressure release, are passed. These preliminary, unoptimised results, which can be seen in Chapter 8, establish the viability of the sol-gel films for DO sensing.

5.2.2 Pulse lifetime system

Fluorescence decay analysis was performed by exciting the sol-gel-coated sensing substrates with 15 ns pulses from a frequency-doubled Nd-YAG laser ($\lambda = 532$ nm). The basic structure of the system seen in Fig. 5.4, is quite similar to the laser-based fluorescence intensity system, both consisting of a Jobin-Yvon 1 metre spectrometer, Hamamatsu photomultiplier tube, C, and pre-amplifier, D. As before, the sensing substrates are mounted in a gas cell and a range of oxygen partial pressures are achieved by precise mixing of oxygen and nitrogen using mass flow controllers and a mixing unit.

From the Stern-Volmer equation, Eqn. 2.13, it can be seen that as the concentration of oxygen increases, the decay time decreases. The fluorescence

decays were not single exponential, due to the lack of homogeneity of the fluorophore environment, and had to be approximated to a double exponential decay as discussed in Section 2.5.

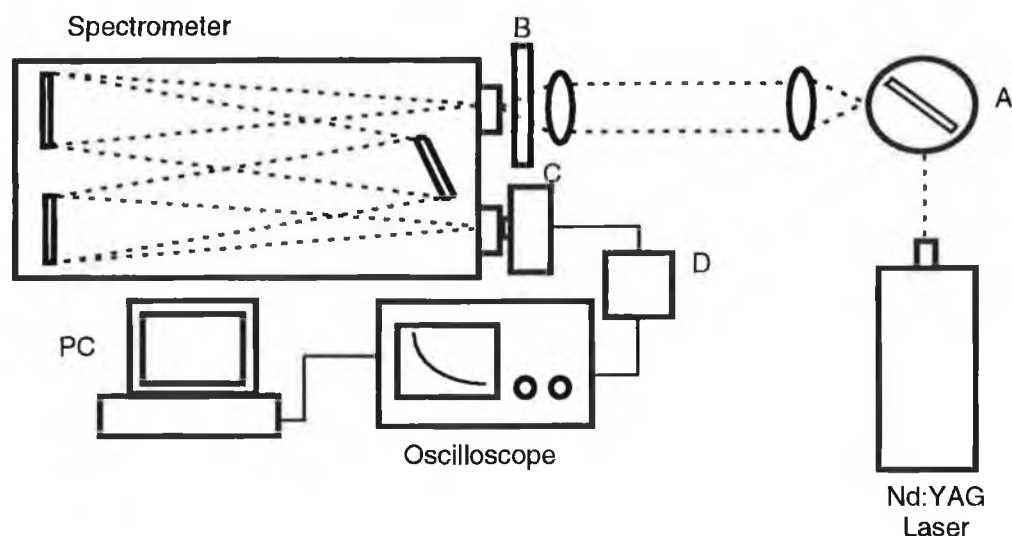


Figure 5.4: System for measuring fluorescence decay times.

Pulse lifetime measurements were carried out in gas phase for TEOS-based films. As will be seen in Chapter 8, these measured lifetime data were used to calculate the expected phase shift for any given oxygen concentration at a particular modulation frequency. They were then compared with the experimental data obtained using phase fluorimetry, and found to be in good agreement.

5.3 LED-based system

The development of LEDs has greatly assisted the evolution of optical sensor technology. Prior to this, lasers and white light sources were the standard excitation sources. In the case of lasers, their large size as well as their power consumption, limits the sensor portability, and the non-selectivity of white light sources leads to inefficient excitation and low intensity of fluorescence. Initially LEDs had quite low output power e.g. initial experiments were carried out with a blue LED having a luminescent intensity of 30 mcd. Their spectral peak occurs at 467 nm, and are fabricated from silicon carbide. Since then, however, ultra bright blue LEDs, fabricated from gallium nitride, and having a luminescent intensity of 1000 mcd, have been made available. Their spectral peak occurs at 450 nm with

output power of 1 mW, and this spectrum is a very good match with the absorption spectrum of the $[\text{Ru}^{\text{III}}(\text{Ph}_2\text{phen})_3]^{2+}$ complex as can be seen in Figure 5.5.

As a result of the increased availability of high output power LEDs, they have become particularly attractive as radiation sources for the following reasons:

- (i) their small size.
- (ii) good optical stability.
- (iii) they are inexpensive.
- (iv) they are easily modulated which enables synchronous detection.
- (v) low power consumption allows the use of a battery.

Consequently, great advances have been made in the field of optical sensors, and LEDs have resulted in the feasibility of miniaturisation and portability of these sensors.

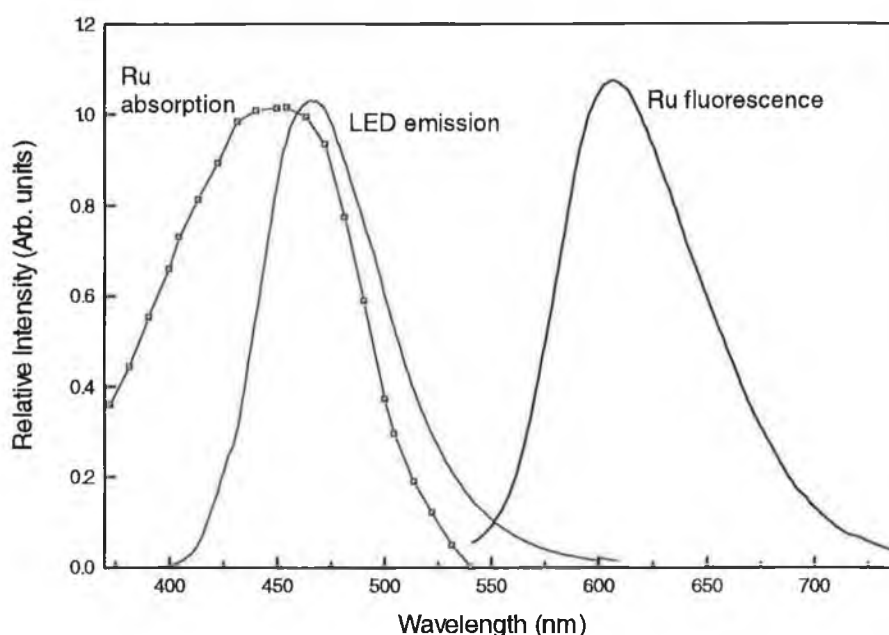


Figure 5.5: Overlap between absorption spectrum of ruthenium complex and emission spectrum of LED.

An all-solid state sensor is described below. The viability of a sol-gel-based sensor for dissolved oxygen was established using the laser-based system described above. Having done this, it was important to design a stable system whereby repeatable experiments could be performed. Planar substrates were used initially, after which a fibre-based system was set-up, demonstrating that a fibre-based sensor configuration was also possible.

5.3.1 Intensity-based planar substrate system

Figure 5.6 shows a schematic of the characterisation system used to measure the response of the sensing substrates in both oxygen gas and dissolved oxygen. The excitation source is a high intensity blue LED (Ledtronics, Torrance, CA.) whose spectral output peaks at 450 nm, and which delivers more than 1 mW of CW optical power. The emission spectrum has good overlap with the absorption spectrum of the ruthenium complex. To prevent overlap between the LED excitation and fluorescence emission, the excitation light passes through a wide band pass filter, 400-505 nm, (Infrared Engineering, Maldon, Essex, U.K.). It is then focused onto a doped sol-gel-coated substrate, which is held just off 45° to the excitation beam in a sealed sample chamber. Fluorescence from the coated substrate passes through a long-wave pass filter, $\lambda_{\text{cut-on}} = 570$ nm, (CVI Laser Corporation, Albuquerque, N.M., U.S.A.) and is focused onto a silicon photodiode detector (Hamamatsu, Middlesex, U.K.; Model S1223-01).

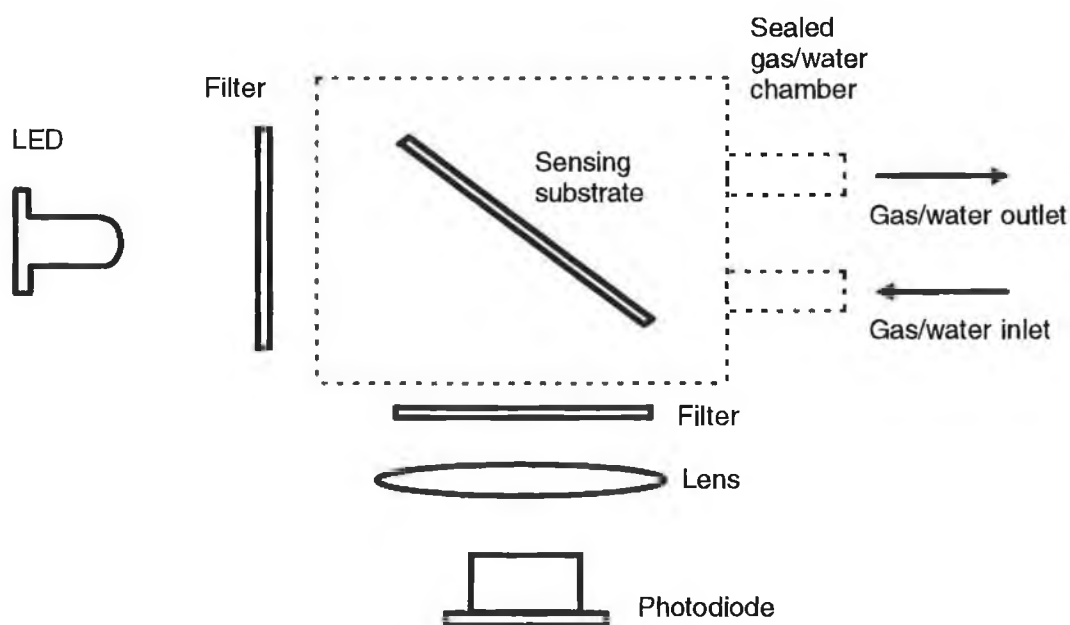


Figure 5.6: Schematic of DO characterisation system.

The filter combination minimises the detection of excitation light with minimal reduction of the fluorescence signal. Synchronous detection with the LED pulsing signal ($f_m \approx 900$ Hz) is achieved by use of an integrated circuit balanced demodulator (Analog Devices; AD 630) which enables effective lock-in detection and discrimination against ambient light.¹ The detection system together with the

ultra-bright blue LED and filter combinations, serves not only to provide a high signal to noise ratio, but allows for miniaturisation and portability. The signal from the photodiode which is digitised by a multifunction input / output board (Bytronics, West Midlands, U.K.; Model MPIBM3), is passed to a PC for analysis and display. For gas phase measurements, oxygen and nitrogen are mixed and flowed via mass flow controllers directly to the sealed gas chamber for measurement, as before. Water samples are prepared by flowing a gas mixture of nitrogen and oxygen from mass flow controllers into a sealed reservoir of water. The gas and water are mixed vigorously using a magnetic stirrer. The sample is then flowed via a peristaltic pump (Gilson, France; Model Minipuls 3) to the water chamber for measurement. All measurements were performed at ambient temperature.

There are three versions of the sample chamber. The first cell, made from aluminium, has inner dimensions of 50 x 30 x 15 mm giving an approximate volume of 23 cm³. There are removable windows on perpendicular sides to allow the sample to be excited and to collect the fluorescence, respectively. On either end of each of the windows, there is a mounting bar so that the LED, photodiode, lenses and filters can be mounted in a stable configuration and moved to facilitate changes in lens focal lengths and distance of the LED / photodiode from the sample. This diagram is shown in Appendix 1(i). Around each window there is an o-ring to prevent leakages. The planar sensing substrate is held rigidly at approximately 45° on the lid of the cell, and the lid is secured tightly to the main body of the cell, again using an o-ring to prevent leakages. There is a hole in the lid of the cell to incorporate an electrochemical DO probe (World Precision Instruments, U.K.; Model ISO₂) for calibration purposes. There are two two-way connectors for flushing the gas/water to be measured through the cell. This cell is very suitable for gas-phase measurements, but due to its large volume it is inefficient for aqueous phase measurements, requiring long gas equilibration and measurement times.

Consequently, a smaller version of the cell was constructed, with inner dimensions 40 mm x 15 mm x 15 mm resulting in a smaller volume of approximately 9 cm³. A diagram of this reduced volume cell is shown in Appendix 1(ii). The lower limit of the cell volume is limited by the size of the lenses and filters (approx. 25 mm in diameter) and because the cell is constructed from aluminium. This reduction in size improved the delivery time of the solutions to the cell and also the scan time. The sensor response time was reduced from over 5 minutes to under 2 minutes. This is not the true response time of the sensing film, but includes the fill time of

the cell which has a major effect on the overall response time in aqueous phase. To minimise this effect, the volume of the cell should be of the order of the size of the sensing substrate. This will be discussed further in Section 7.3.

To try to attain this, a 1 cm^3 volume cell was designed, which was fabricated from glass by a specialist company (Optiglass, Essex, U.K.). This allows us to retain the original design while reducing the cell volume. The basic structure of the cell, which can be seen in Appendix 1(iii), consists of a rectangular glass cell with a diagonal slit ultrasonically drilled out of the centre. There are two tubes, the lower being the inlet feeding the slot with water, and the upper being the outlet by which water is pumped from the slot.

Special planar glass substrates were cut to size with a hole in the top for mounting on the lid. The cell lid, which is also illustrated in Appendix 1(iii), then bolts down onto the main cell body. Because the cell material is optical glass, both excitation and fluorescence light can pass through it, but the actual capacity of the cell for liquid is very small. This results in response times which are closer to the true response time of the film, rather than being limited by the fill time of the cell, as previously seen.

5.3.2 Intensity-based fibre substrate system

The basic experimental system was extended to develop an evanescent-wave fibre optic sensor, which is shown in Figure 5.7. The optical fibre used was that described in Section 4.3, plastic clad silica fibre with core diameter of $600\text{ }\mu\text{m}$.

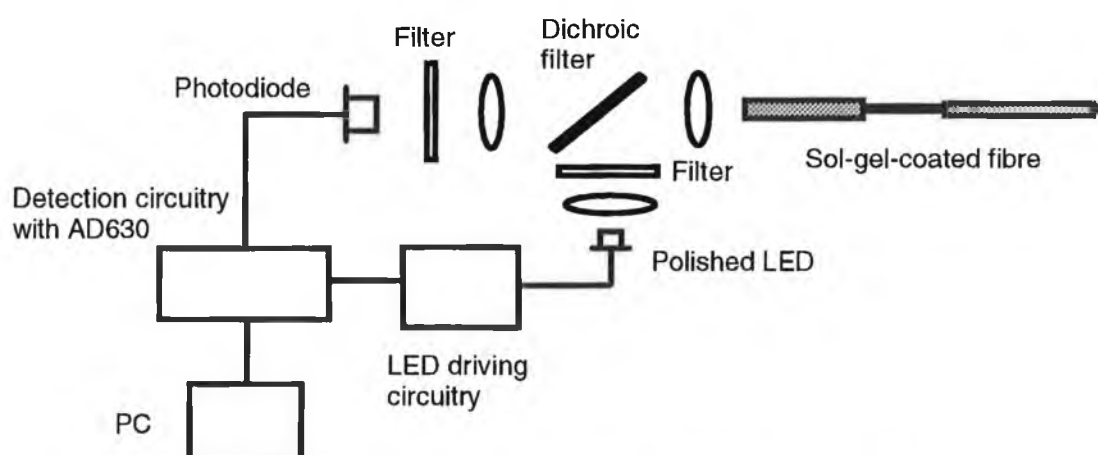


Figure 5.7: Schematic of fibre-based fluorescence intensity sensor.

As before, the sol-gel-coated fibre was excited using an ultra-bright blue LED, but in this case the dome was polished down to a level close to the emitting surface, to achieve optimum launching.² The excitation light passed through a 10 mm lens, and then through a wide band pass filter, after which it was incident on a dichroic colour separation filter. This filter (Reynard Corporation, San Clemente, CA., U.S.A.) reflects the incident blue light into the microscope objective lens and into the optical fibre. The resultant fluorescence is then transmitted by the dichroic filter and focused onto the photodiode.

There are many advantages to collecting the fluorescence from the proximal end of the fibre, i.e. the end of the fibre directly excited by the excitation source, among which are: the SNR is greater for proximal end than for distal end collection, as of the order of 90% of the excitation light that is launched exits at the distal end, the end furthest from the excitation source; it allows the use of long fibres for remote sensing; and this configuration makes installation and alignment easier.

The fibre was mounted in a flow cell, and was held in place using septa, hence water was pumped through the cell without movement of the fibre. All the components were mounted on a 4-bar rail, whereby linear adjustment was easily achieved.

5.3.3 Phase fluorimetry system

The system used for phase fluorimetric measurements is shown in Figure 5.8. The ultra-bright blue LED is sinusoidally modulated using a function generator at an optimum modulation frequency.

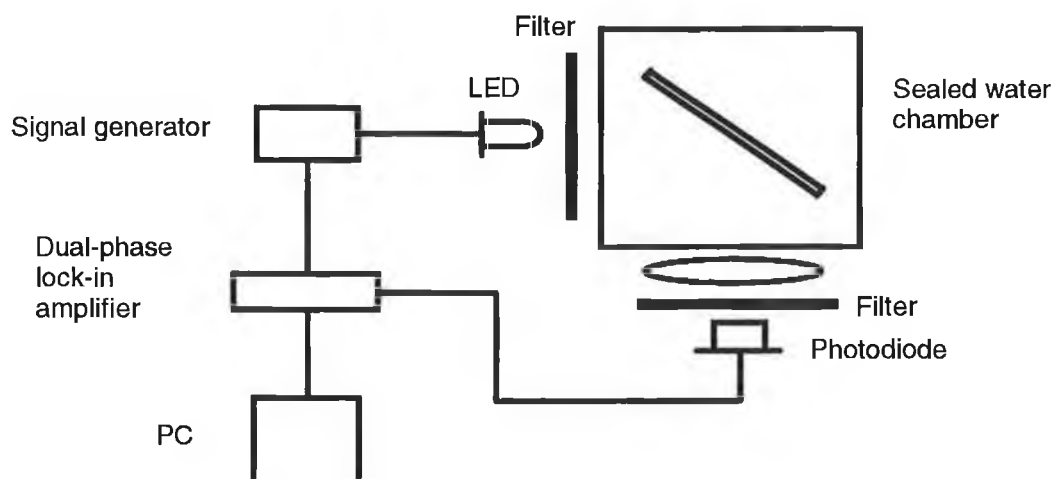


Figure 5.8: Schematic of phase fluorimetric DO sensing system.

The optimisation of the modulation frequency is detailed in Chapter 8. The optimum modulation frequency was found to be 45 kHz. In the detection circuitry, it was necessary to use a low bias current operational amplifier to amplify the signal detected by the photodiode. The OPA-128 (Burr Brown, Tuscon, AZ., U.S.A.) allows amplification of 10^8 and has an ultra low bias current of 75 fA.³ A dual-phase lock-in amplifier (EG&G, Model 5210) was used to measure the phase angle at each oxygen concentration.

The LED modulation frequency was used as reference. The output from the dual-phase lock-in amplifier passes to the PC via the multifunction input/output board, and is saved in a datafile for analysis. The response of this experimental system to dissolved oxygen is discussed in Chapter 8.

5.4 System for obtaining calibrated DO solutions

This is a simplified version of the instrumentation used to obtain tonometered liquids by Joanneum Research.⁴ They have a number of cylinders of accurate mixtures of nitrogen and oxygen gas, each of which are bubbled into separate sealed gas washing bottles. The delivery of these solutions to the oxygen detector is controlled by a valve-switching system and a peristaltic pump.

The system used in this work is less elaborate, but quite accurate, and it can be seen in Fig. 5.9.

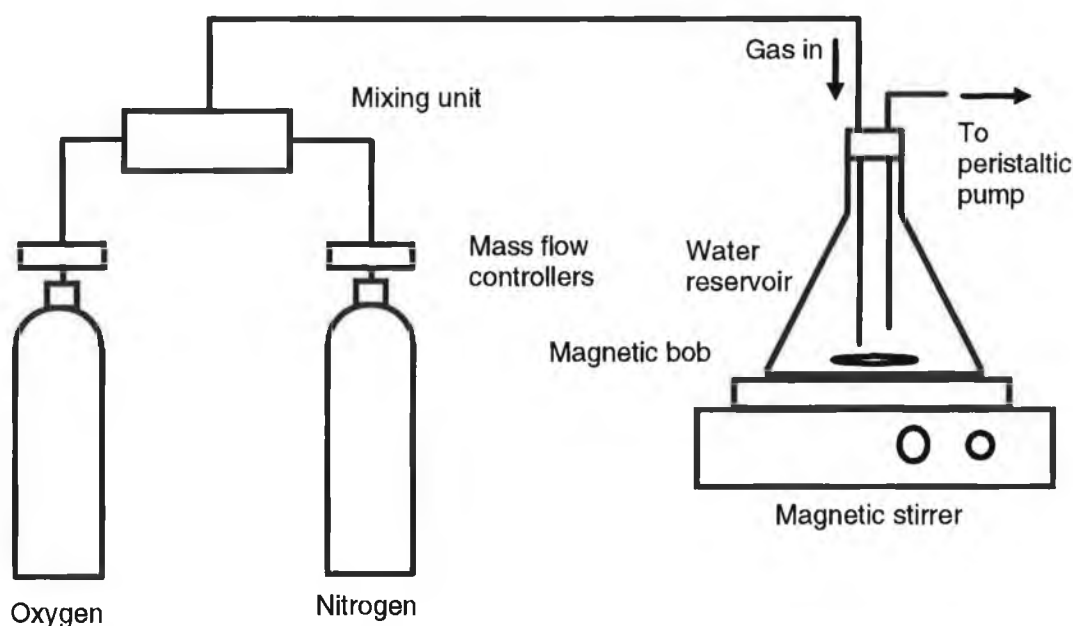


Figure 5.9: System for obtaining calibrated DO solutions.

It consists of two cylinders of medical grade nitrogen and oxygen gas, whose flow is controlled by mass flow controllers. Depending on the concentration of DO required, the ratios of gases can be adjusted accordingly. The gas mixture was flowed into a sealed reservoir of water (deionised water was generally used so that repeatable calibrated solutions could be obtained). The reservoir contains a magnetic bob, and is positioned on a magnetic stirrer. The calibrated solution is obtained by vigorous stirring, such that the agitation accelerates the diffusion of gases through the water, resulting in equilibrated solutions.

The solutions of dissolved oxygen obtained using this system can be calibrated using a Clark electrode (ISO₂ dissolved oxygen probe), after which they are pumped to the characterisation cell for measurement using a peristaltic pump. In the case of low concentrations of DO, the system is changed slightly, incorporating a low flow oxygen mass flow controller (0 to 10 sccm), and rearranging the system such that the peristaltic pump is placed after the cell to pull the solutions through from the reservoir. The low concentration DO solutions flow directly to the cell via narrow bore stainless steel tubing to avoid any gas exchange between the solutions in the PVC tubing and the external atmosphere.

5.5 Humid atmosphere test system

Figures 5.10(i) and (ii) show systems for investigating the effect of highly humid gas on the sol-gel sensor. Preliminary experiments were performed using Fig. 5.10(i).

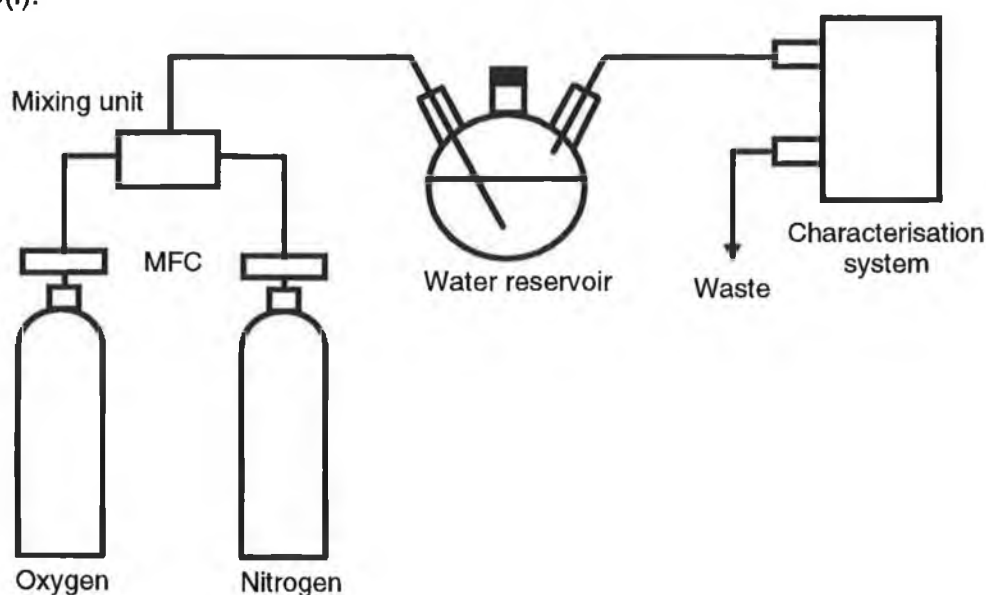


Figure 5.10(i): Initial humid gas test system.

Fig. 5.10(ii) shows the second and more accurate system for obtaining fully humid gas. The gas mixture from the mass flow controllers flows into the first gas washing bottle. The gas adsorbs some moisture and flows through the second gas-washing bottle. Each time the gas adsorbs more and more moisture, until it is finally fully humidified. The response of the sensor to humidified gas is compared with that of ambient gas, by switching the valve between the two pathways. A temperature / humidity probe (Eirelec; Model TH-210) is incorporated into the set-up after the final gas-washing bottle to measure the relative humidity. This set-up achieved gas with a relative humidity of 99.8%. The results from these humidified gas experiments are shown in Section 7.4.

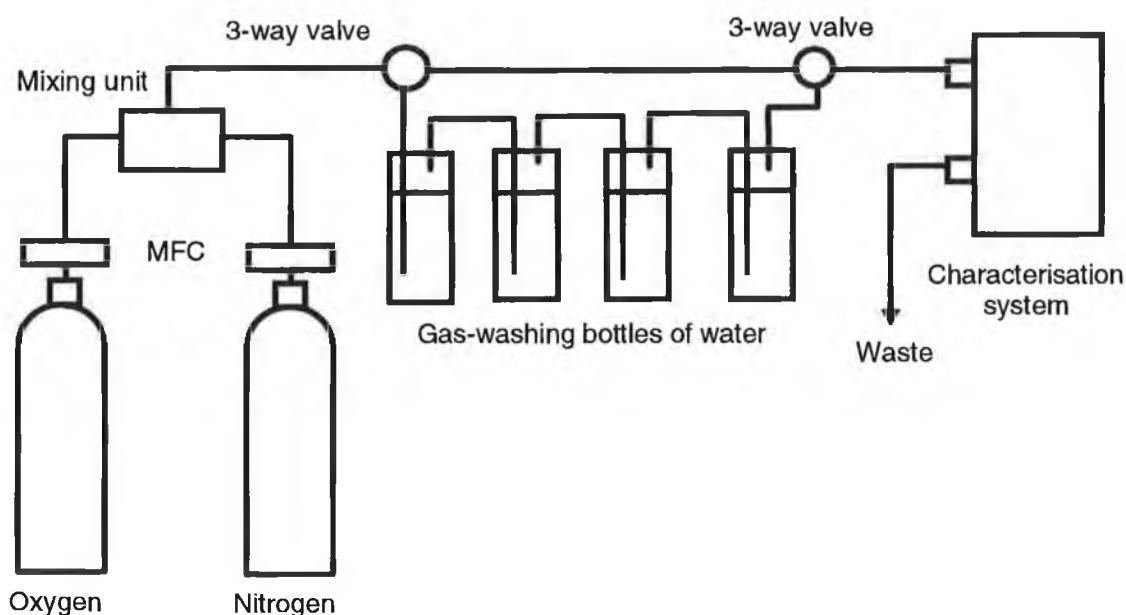


Figure 5.10(ii): System for humidifying gases

5.6 Interference test system

An interference test system was assembled in order to investigate the effect of contaminants which may be present in test water, on the response of the sensor. This system is illustrated in Figure 5.11. It consists of two reservoirs, one containing uncontaminated reference water, and the second containing the interferent solution to be tested. Both of the reservoirs are exposed to ambient atmospheric pressure and temperature. When the solutions have equilibrated, the uncontaminated water is pumped into the cell for measurement, achieving a

reference intensity level. Having reached a steady-state intensity reading, the interferent solution is pumped into the cell for measurement. A drop in intensity between the two levels suggests that some sort of quenching or interference is taking place, which can be quantified.

Interference tests are not trivial experiments to perform. Discrepancies may be caused by substances which, by their nature, consume oxygen, for example algae or bacteria, or substances which change the oxygen solubility and hence the DO concentration, by changing the pH of the water. Substances such as sodium sulphite and sodium hydroxide change the DO concentration.

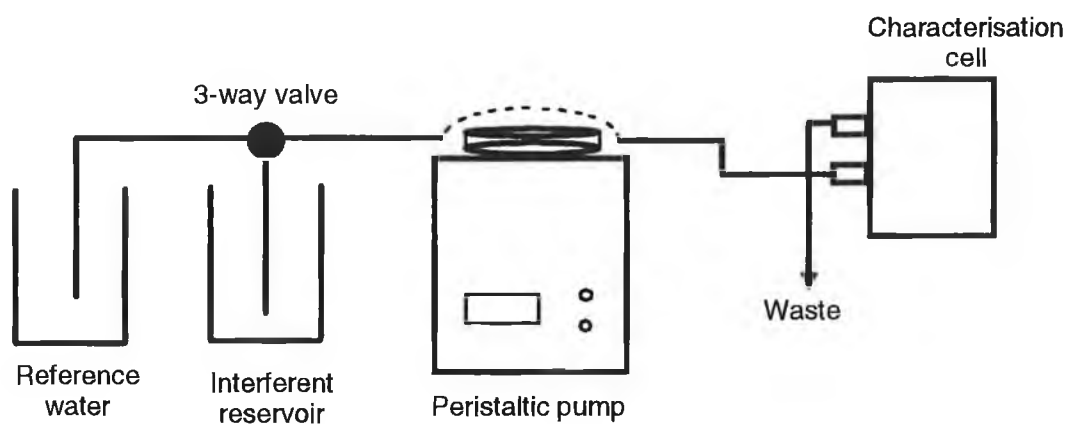


Figure 5.11: Interference test system.

In this case, it is necessary to use an independent measurement source, the Clark electrode, to monitor the DO concentration of the solutions.

A second set of interference tests were carried out using nitrogen-purged solutions.⁵ More conclusive results were obtained in this case as the nitrogen-purged solutions help to eliminate any problems caused by any differences in DO concentrations between the two reservoirs.

Interferents tested include heavy metals, surfactants, and tap water. Teflon tubing is substituted with stainless steel tubing when testing the effect of surfactants on the sensor, as surfactants coat the inside of Teflon and PVC tubing.

5.7 Summary

The experimental systems used in this work have been described in this chapter. Both preliminary laser-based fluorescence intensity and lifetime systems as well as miniature all-solid state systems, both planar- and optical fibre-based, have been detailed. A phase fluorimetric sensor has also been outlined. Systems for characterising the sensors under certain conditions have been illustrated, such as the interferent test system, the system for obtaining calibrated DO samples, and the gas humidifying system. Results from these systems are shown in Chapters 6, 7, and 8.

References:

1. P. C. Hauser, S. S. Tan, **All-solid state instrument for fluorescence-based fibre optic sensors**, *Analyst*, 1993, Vol. 118, pp. 991-995
2. G. O'Keeffe, **Development of fibre-optic evanescent-wave fluorescence-based sensors**, PhD thesis, 1995, Dublin City University
3. **Difet electrometer-grade operational amplifier** - Data sheet OPA128, Burr Brown, Tuscon, AZ., U.S.A.
4. **Optical Oxygen Sensor: calibration**; Joanneum Research ISI, Graz, Austria, 1993.
5. I. Klimant, O. S. Wolfbeis; **Oxygen-Sensitive Luminescent Materials Based on Silicone-Soluble Ruthenium Diimine Complexes**, *Anal. Chem.*, 1995, Vol. 67, No. 18, pp. 3160-3166

CHAPTER 6: OPTIMISATION OF SENSING FILMS

6.1 Introduction

This chapter outlines the progress made in film optimisation for dissolved oxygen sensors. Earlier studies of TEOS-based films established the viability of ruthenium-doped sol-gel films for oxygen sensing. For dissolved oxygen sensing, much preliminary work was also carried out on TEOS-based films. It is shown here that the microstructure can be tailored to optimise the quenching response of the sensor in particular oxygen gas concentration ranges. The response of TEOS-based sensors was investigated in aqueous phase, and they were found to have a much lower quenching response than in gas phase. This was thought to be as a result of the hydrophilic nature of the sol-gel film surface, which allows the adsorption of water onto the surface¹, thus decreasing the effective pore size. Consequently, the film surface was modified to eliminate this effect by using organically modified precursors in the fabrication procedure. Optimisation of the organically modified films for dissolved oxygen sensing is traced, and quenching behaviour of films in both gas and DO phase is discussed.

6.2 TEOS-based films

Tetraethoxysilane (TEOS) is a silicon alkoxide precursor, consisting of a central silicon metal ion, which is surrounded by four ethoxy ($-\text{OC}_2\text{H}_5$) groups. Initial quenching experiments were performed on the oxygen-sensitive ruthenium complex dissolved in ethanol, and these confirmed the high sensitivity of the complex to oxygen gas. A sol-gel film was the chosen immobilisation matrix for the dye complex due to its obvious advantages over other immobilisation methods² (See Section 1.5). Porous TEOS-based films dried at 70°C, to retain the porosity and prevent thermal degradation of the dye complex, were used as a support for the oxygen-sensitive dye.

The following sub-sections deal with the oxygen response of TEOS-based films, both in gas phase and aqueous phase, and the effect of microstructural variations on the response of these films.

6.2.1 Gas phase measurements

Gas phase measurements were carried out using the set-up illustrated in Figure 5.3. Quenching experiments have been used as a tool to characterise the films as a function of the changing microstructure. In these experiments, no allowance has been made for the effect of humidity on the results. Ambient humidity has been assumed in all experiments. As will be seen later, humidity has a sizeable effect on the quenching response for gas-phase measurements. The quenching data demonstrates how, by varying some of the process parameters, the response can be tailored for different applications. It has also been demonstrated that the quenching response in gas phase is independent of thickness for thicknesses ranging from 360 nm to 450 nm.

6.2.1.1 Quenching behaviour

The quenching behaviour of a typical TEOS film can be seen in Figure 6.1. These films were fabricated at pH=1 and R=2, which gives a microporous film suitable for oxygen quenching. This structure entraps the dye complex, and prevents dye leaching, while retaining the accessibility of the analyte. The films were exposed to alternate environments of 100% oxygen and 100% nitrogen gas and the response was monitored and recorded.

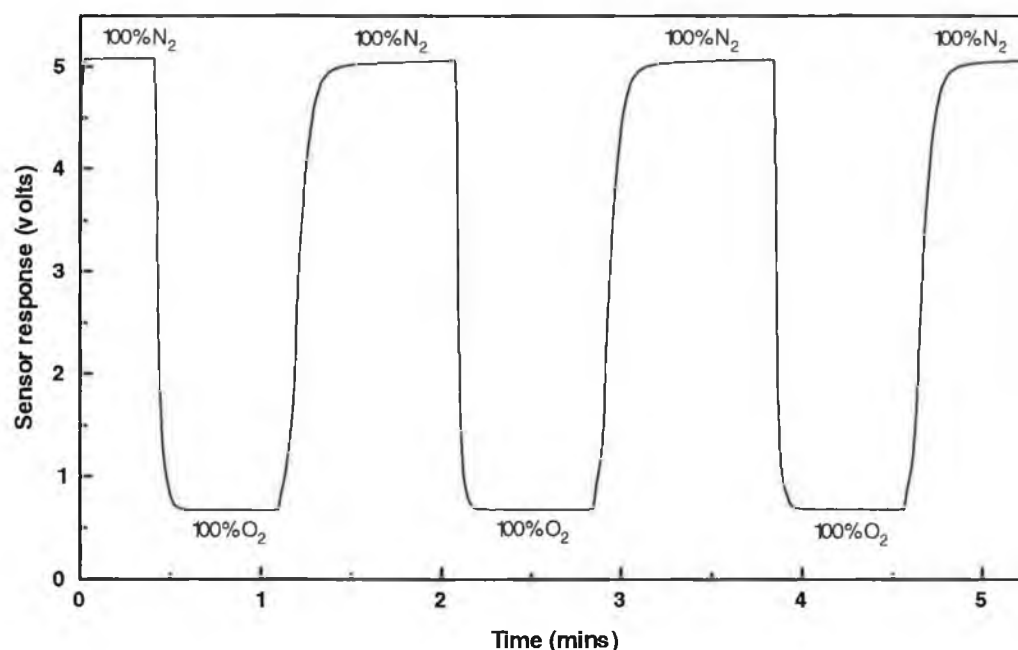


Figure 6.1: Response of R=2 TEOS film to 100% N₂ and 100% O₂ gas.

The overall quenching response in gas phase is denoted by Q_G , and defined as follows,

$$Q_G = \frac{I_{N_2} - I_{O_2}}{I_{N_2}} \cdot 100 \quad (6.1)$$

where I_{N_2} is the fluorescence intensity in 100% nitrogen gas, and I_{O_2} is the fluorescence intensity in 100% oxygen gas. For the TEOS films, Q_G is $89 \pm 1\%$, where 1% is the standard deviation of 5 readings. This signifies a high level of accessibility of the ruthenium dye complex in the sol-gel matrix by the oxygen gas. A high level of repeatability and excellent reversibility are characteristic of the response of TEOS films in gas phase, as shown in Fig. 6.1. Furthermore, the quenching data exhibit a high signal-to-noise ratio.

6.2.1.2 Tailoring the microstructure

The microstructure of the sol-gel film is highly dependent on the process parameters and on the fabrication procedure, as was detailed in Chapter 3. In this work, the R value (ratio of water to precursor) was varied to investigate the effect on quenching and other parameters such as stabilisation time of the films. According to Eqn. 3.4, an R value of 2 is theoretically sufficient for complete hydrolysis and condensation.³ However, even in excess water, $R > 2$, the reaction does not go to completion. As will be seen in Section 6.2.3, this has a knock-on effect, in that the stabilisation of the microstructure of the film is not immediate. Here, the response of the films, fabricated with R values of 2, 3 and 4, to different concentrations of oxygen is examined. Table 6.1 shows the percentage quenching of R=2, R=3 and R=4 TEOS films in 10% oxygen and 100% oxygen. These data give an indication of the microstructural differences between the films.

	R=2 TEOS	R=3 TEOS	R=4 TEOS
Q_{10}	39%	26%	22%
Q_{100}	88%	86%	83%

Table 6.1: Quenching response in 10% and 100% oxygen for R=2, 3 and 4 TEOS films.

Figures 6.2, 6.3 and 6.4 show the gas phase calibration data, i.e. graphs of fluorescence intensity as a function of oxygen gas concentration, of R=2, R=3 and

R=4 films. As can be seen, the differences between these plots are quite significant.

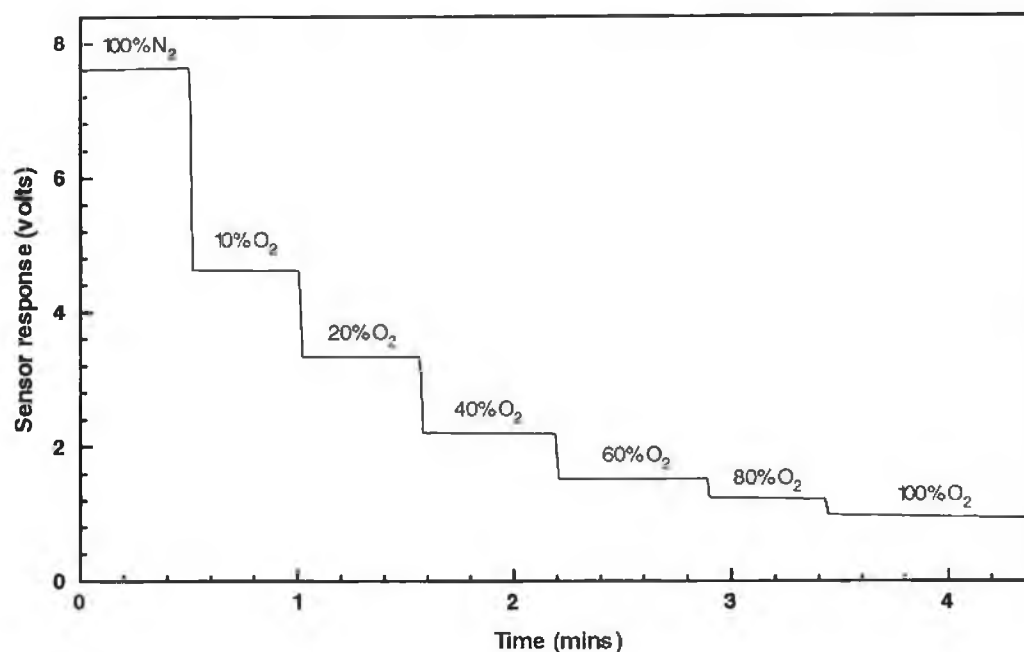


Figure 6.2: Gas phase calibration data for R=2 TEOS film.

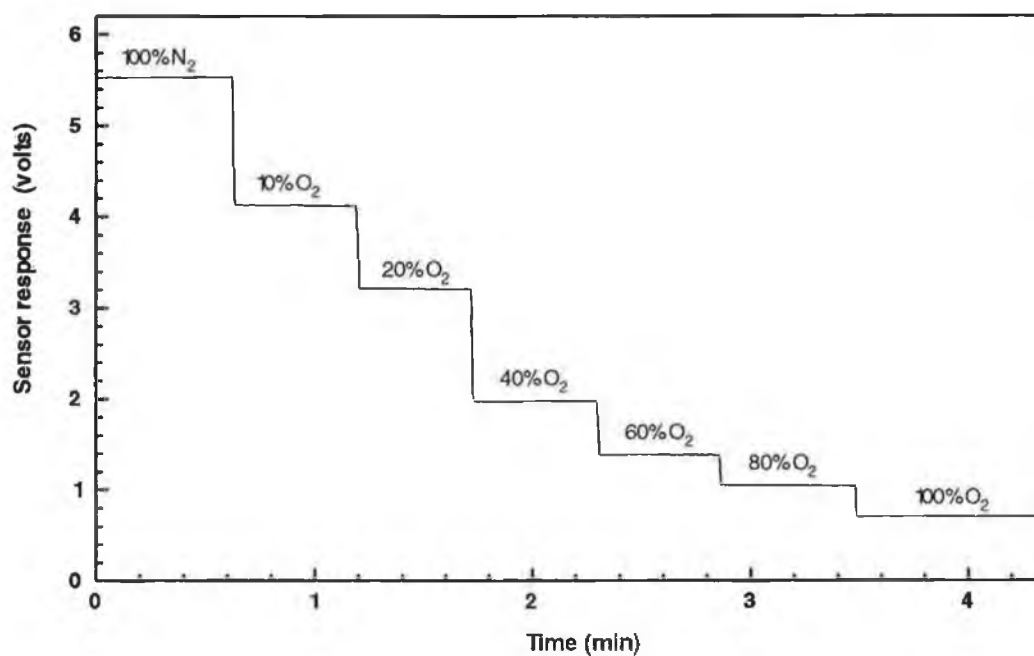


Figure 6.3: Gas phase calibration data for R=3 TEOS film.

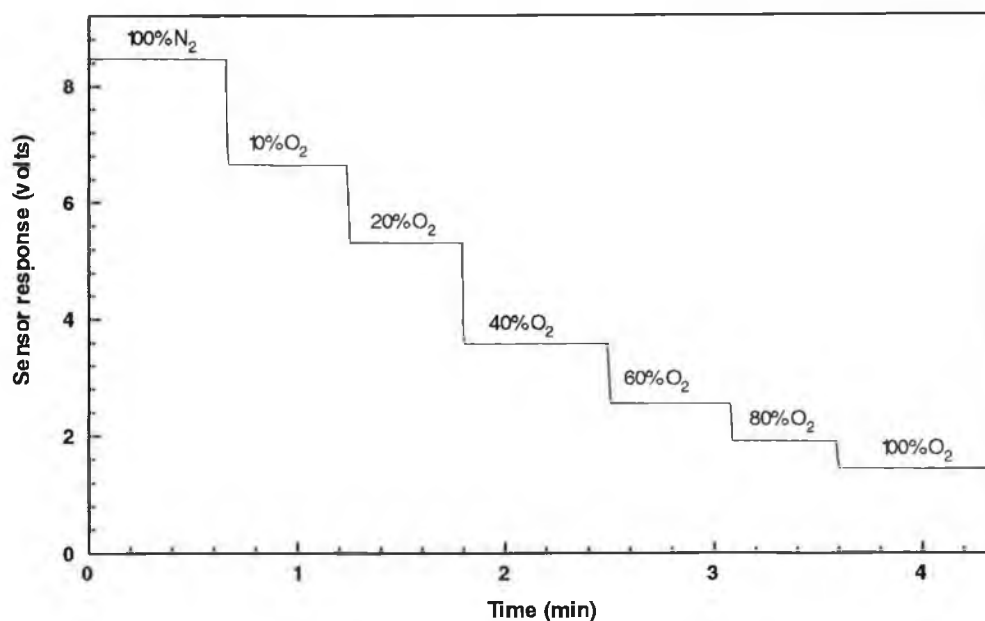


Figure 6.4: Calibration data for R=4 TEOS film in oxygen gas..

For R=2 films, the response is highly non-linear and the film shows a high sensitivity to oxygen at low oxygen concentrations. For films with R=4, however, the response has a smaller rate of change of intensity at low oxygen concentrations. For R=3 films, the response is intermediate between that for R=2 and R=4 films. Thus, by varying the R value, films can be tailored to redistribute the quenching response in a particular concentration range. The Stern-Volmer relationship, according to Eqn. 2.12, is plotted in Fig. 6.5 for R=2 and R=4 TEOS films. Note that Q_G increases with K_{SV} , the slope of the Stern-Volmer plot. From the figures it is clear that the largest Q_G and K_{SV} values (R=2 films) correspond to the largest intensity rate of change. From Eqns. 2.4 and 2.5, K_{SV} is proportional to k , the bimolecular quenching constant, which in turn is proportional to D , the oxygen diffusion coefficient in the film. For gas-phase sensing, the diffusion process is pore-size limited.⁴ Hence these results indicate that the R=2 films have a larger average pore size, resulting in a larger value of D than R=4 films. This is consistent with a slightly denser more cross-linked microstructure for R=4 films arising from faster hydrolysis and condensation rates due to increased water content. This conclusion is corroborated by leaching studies recently performed in this laboratory using the pH sensitive dye, bromophenol blue (BPB) which is smaller than our Ru complex (11 Angstroms as compared with 25 Angstroms). It

was found that the dye leaching rate from R=2 films was considerably faster than for R=4 films.⁵ It is assumed here that for gas phase sensing, the solubility, α , in Eqn. 2.5 is constant for the films and that changes in the microstructure affect only the diffusion behaviour. This is not the case for quenching in DO as discussed in Section 6.3. Finally, although no direct methods of pore size characterisation have been carried out in this laboratory, it is clear that indirect estimates of relative pore size as a function of processing parameters can be deduced from oxygen quenching behaviour and leaching experiments.

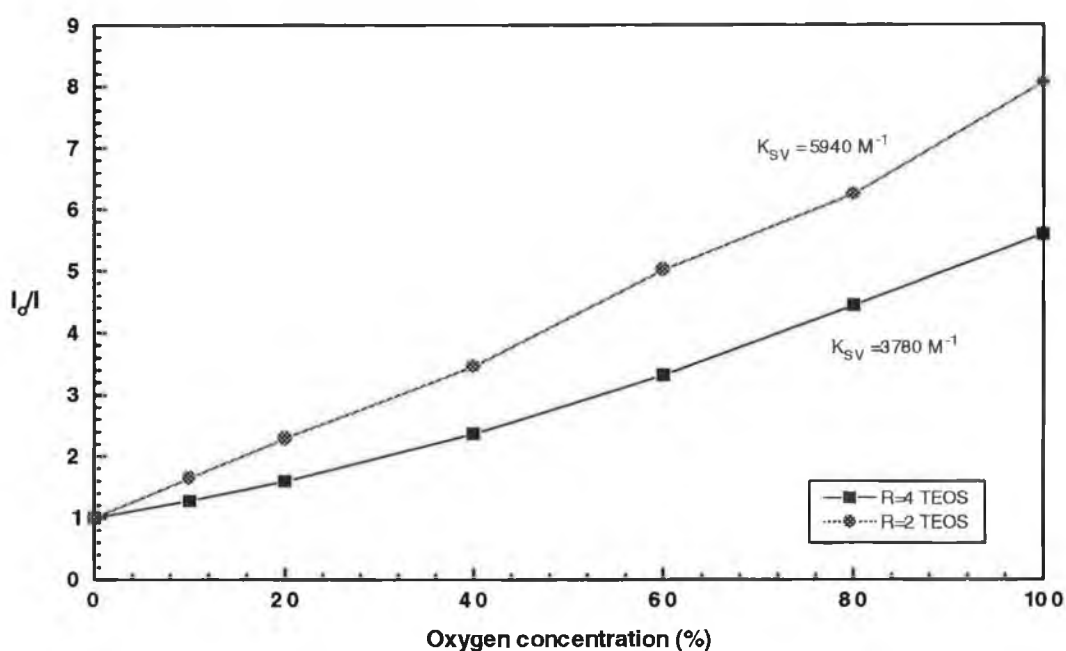


Figure 6.5: Stern-Volmer plots of R=2 and R=4 TEOS films in oxygen gas.

6.2.1.3 Quenching response as a function of thickness

Variations in the amount of water to precursor in sol fabrication cause a change in the viscosity of the sol. According to Eqn. 3.5, the thickness is directly proportional to the viscosity of the coating sol. In order to eliminate the possibility that the variation in film thickness rather than R value caused the change in quenching response, the following investigation was carried out. Films of the same water:TEOS ratio, $R = 4$, were prepared at three different dip speeds, yielding films of thickness 350 nm, 400 nm and 460 nm respectively. The films were dip-coated onto both silicon and glass substrates in order that thickness measurements could

be carried out using the ellipsometer, as well as quenching experiments. The gas phase quenching response for each of the three films can be seen in Figs. 6.6, 6.7 and 6.8.

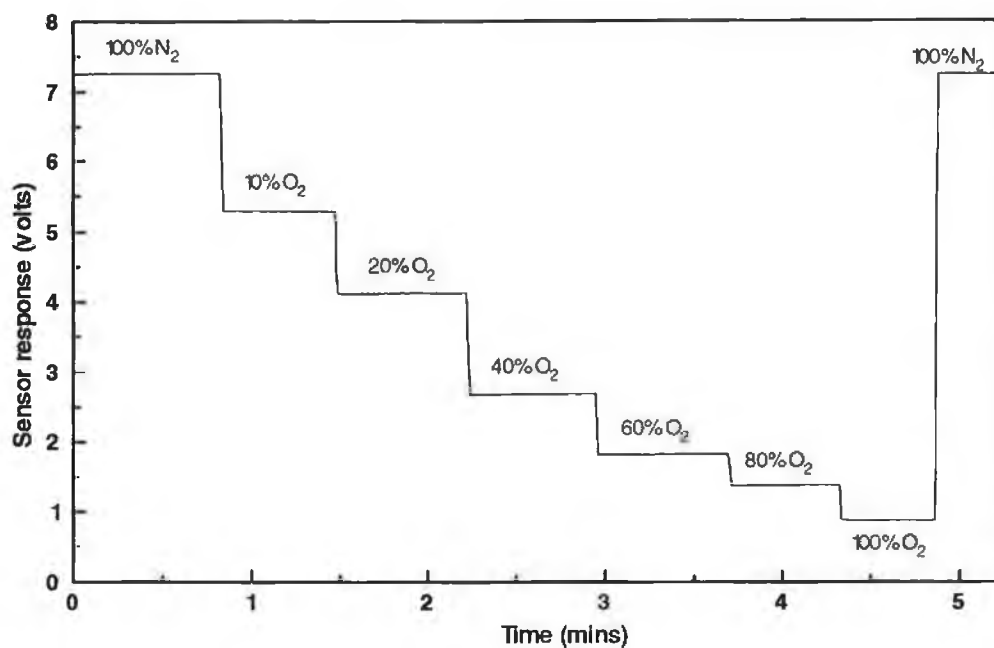


Figure 6.6: Quenching response of R=4 films with film thickness of 350 nm.

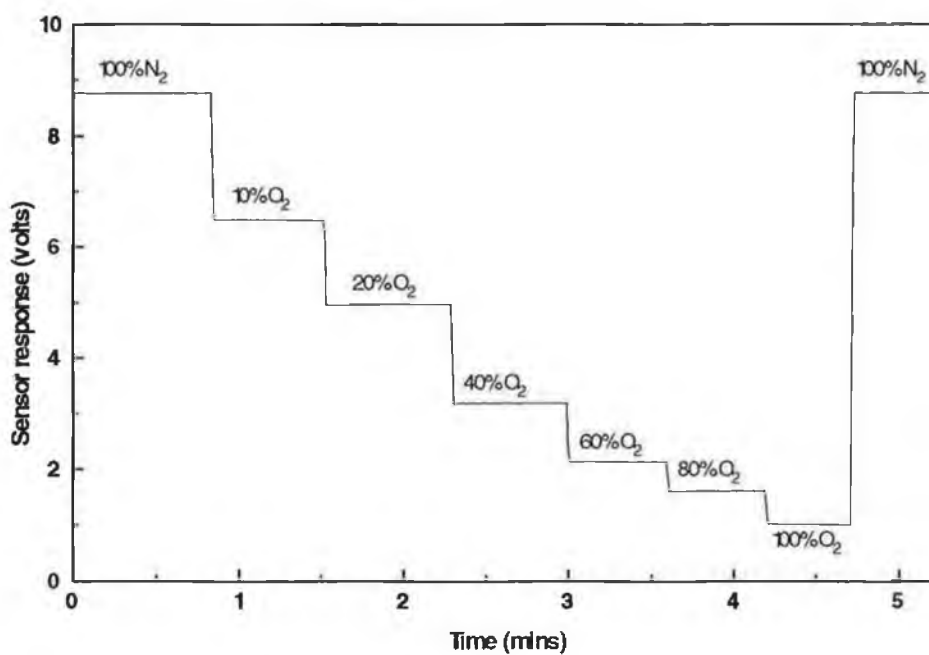


Figure 6.7: Calibration data for R=4 films with film thickness of 400 nm.

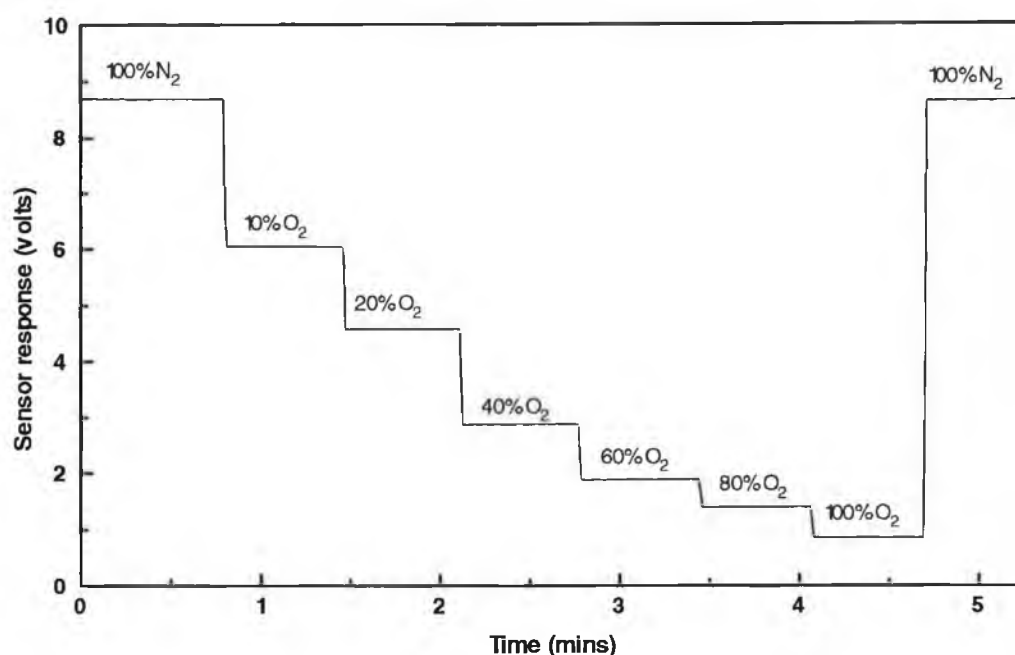


Figure 6.8: Calibration data for R=4 TEOS films with film thickness of 460 nm.

From the graphs it can be seen that the quenching responses are almost identical for the three film thicknesses ($Q_G = 88\%$, 88.4% and 89% respectively), thus indicating that quenching response is independent of film thickness within a particular thickness range. A sizeable change in thickness would however be expected to affect response time. The thicknesses chosen here are within the range used for our sensors.

6.2.2 Aqueous phase measurements

The characterisation systems used for aqueous phase measurements are those shown in Appendix 1(i) and 1(ii). Calibrated DO solutions for aqueous phase measurements were made using the system shown in Fig. 5.9. The solutions were pumped via a peristaltic pump to the characterisation cell for measurement. In this section, the quenching behaviour of the TEOS-based films in aqueous phase is discussed and compared with those in gas phase.

6.2.2.1 Quenching response

The overall quenching response in aqueous phase, Q_{DO} , is defined as follows:

$$Q_{DO} = \frac{I_{deoxy} - I_{oxy}}{I_{deoxy}} \cdot 100 \quad (6.2)$$

where I_{deoxy} is the fluorescence intensity in fully nitrogenated aqueous solution and I_{oxy} is the fluorescence intensity in fully oxygenated aqueous solutions. The error for measurements in aqueous phase is $\pm 2\%$, which represents the standard deviation of five readings.

Preliminary aqueous phase measurements on R=2 TEOS films were encouraging as the fluorescence intensity was shown to quench in DO. However, as can be seen from Fig. 6.9, although the degree of quenching in fully oxygenated water is constant, the intensity is gradually decreasing.

Ongoing characterisation studies led to the discovery that the thickness of these films was not stabilised at the time of measurement (see Section 6.2.3). Another reason for the intensity fall-off was photobleaching due to the high intensity of the blue LED. When the power from the LED was reduced, this effect did not occur, and the high signal-to-noise ratio was maintained. Total oxygenation and deoxygenation was also found to be an important factor in ensuring reversibility of the sensor measurements. Figures 6.10 and 6.11 show the quenching response of stabilised R=2 and R=4 TEOS films.

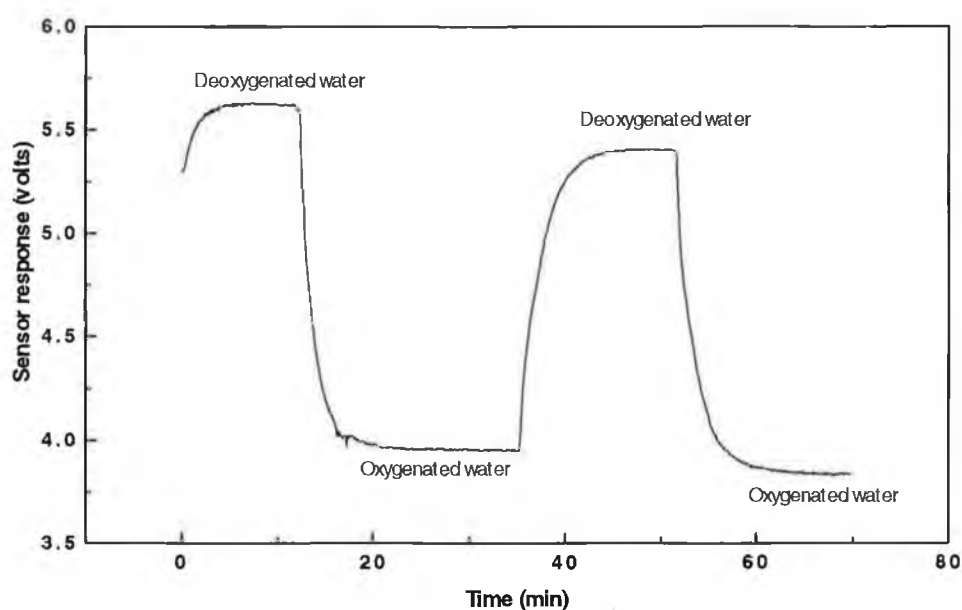


Figure 6.9: Preliminary quenching response of R=2 TEOS films in aqueous phase.

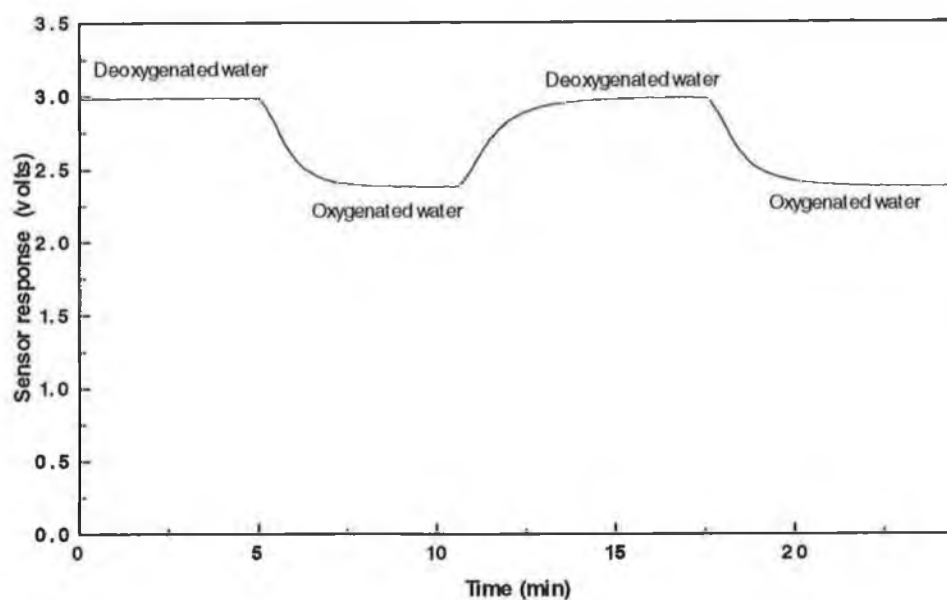


Figure 6.10: Quenching response of stabilised R=2 TEOS films in aqueous phase.

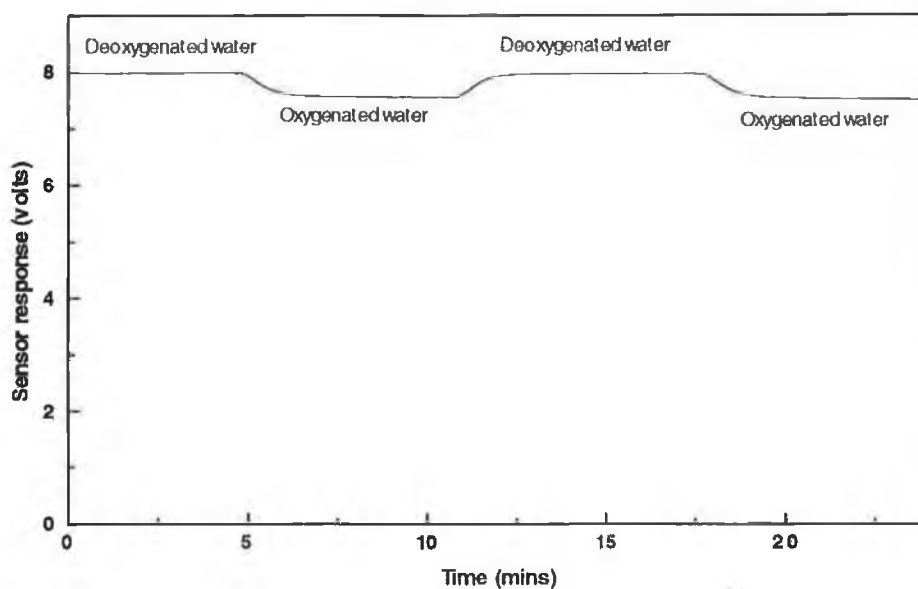


Figure 6.11: Quenching response of stabilised R=4 TEOS films in aqueous phase.

The quenching response of R=2 TEOS films in aqueous phase, Q_{DO} , is $20\% \pm 2\%$, whereas Q_{DO} for R=4 TEOS films is $6\% \pm 2\%$. The most widely used DO sensor, the Clark electrode, operates on the principle whereby oxygen in the water comes

out of solution, passes through a hydrophobic membrane and is detected at the electrode.⁶ In the sol-gel sensors discussed here, it is proposed that a similar principle applies i.e. that the oxygen partitions out of the water at the film / water boundary and accesses the ruthenium complex in the gas phase. It is proposed that the low values of Q_{DO} for TEOS films, relative to the Q_G values already discussed, is due to the relatively small partitioning effect at the water/film boundary i.e. the surface is hydrophilic. As was stated in Chapter 3, the surface of TEOS films is covered with silanol groups which encourage the adsorption of physisorbed water. It is thought that due to this predominantly hydrophilic surface, only a small degree of partitioning from aqueous phase to gas phase occurs. This, along with the fact that quenching by oxygen in dissolved phase is also occurring, accounts for the relatively low quenching response of TEOS-based films in aqueous phase compared with that in gas phase. For a viable DO sensor, a high overall quenching response is necessary to attain good resolution at low oxygen concentrations. Section 6.3 describes how the surface was modified to remove the silanol groups thus increasing the film hydrophobicity and improving the quenching response in water.

6.2.3 Thickness stabilisation

In Section 6.2.2.1, stabilisation of the microstructure was mentioned. Stabilisation of the microstructure refers to the time it takes for the film properties to reach a steady state condition, such that the film thickness or porosity do not change with time. As stated previously, an R value of 2 is the stoichiometric ratio whereby the hydrolysis reaction proceeds to completion, in theory. In practice, due to reverse reactions and other competing processes, R values greater than 2 are required to ensure that hydrolysis is complete before condensation and film drying occur. TEOS films with R values of 2 and 4 were dip-coated and dried at 70°C for 18 h. After drying, the film thicknesses were measured using ellipsometry techniques. The films were stored in ambient conditions, and their thicknesses measured periodically. Fig. 6.12 shows film thickness for each of the films as a function of time.⁷ For R=2 films, the film thickness decreased gradually over approximately 80 days, while for R=4 films, their thickness stabilised in approximately 20 days. These effects are due to incomplete hydrolysis prior to film coating and drying. It is clear from the literature that, even at relatively high R values, unhydrolysed

monomers can be present in the gel.³ As a result, hydrolysis continues during and after the drying process giving rise to a changing microstructure which causes a gradual decrease in film thickness. This continuing hydrolysis is facilitated by exposure to moisture in the atmosphere. An investigation into this proposition was carried out in this laboratory,⁷ and involved measurement of thicknesses of R=2 films which had been stored in a desiccator after drying. These measurements were compared with those of films stored under ambient conditions. The thickness of the former remained approximately constant. By subsequently storing these samples under ambient conditions, thicknesses began to fall as they had done with the latter samples. These measurements are consistent with the theory of continuing hydrolysis in the presence of atmospheric moisture.

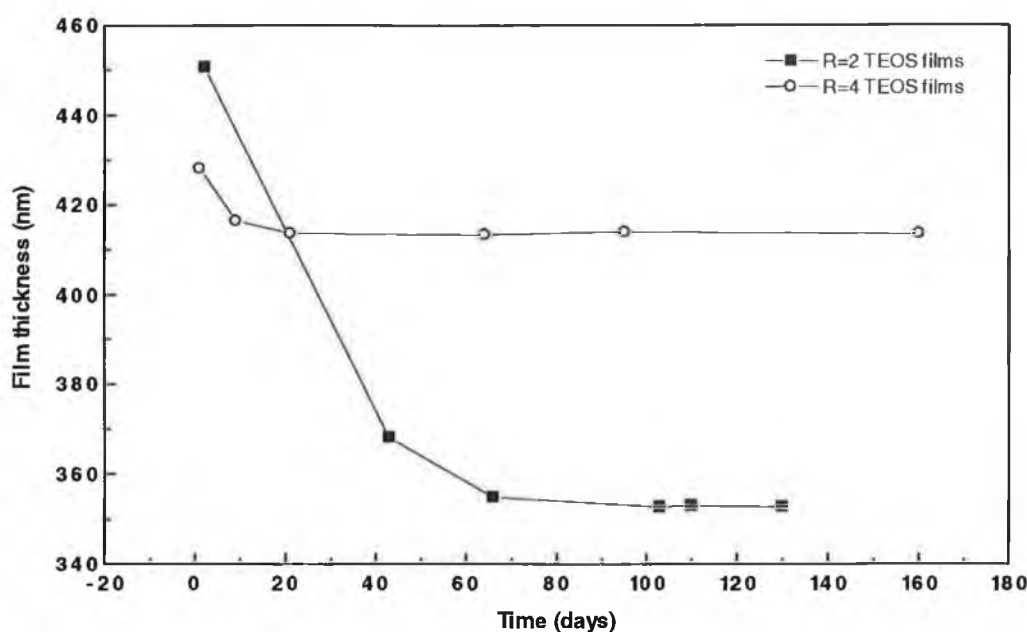


Figure 6.12: Temporal stability of R=2 and R=4 TEOS film thickness

6.2.4 Leaching

A number of investigations were carried out to study the effect of long-term immersion in both static and agitated water. Glass substrates were coated with R=2 and R=4 sols, and these were dried at 70°C. Their respective absorption spectra were measured and they were then immersed in static water. The peak of the absorption spectrum of the ruthenium dye complex at 460 nm was used as a measure of the ruthenium dye concentration. The slides were scanned

periodically using a uv-vis spectrophotometer (Shimadzu; Model UV-1201). Initial measurements were taken every four hours to monitor the initial response. After only 10 hours the 2:1 films showed an overall decrease in absorbance at 460 nm, and this decrease continued, as can be seen in Fig. 6.13. (The initial increase in Fig. 6.13 is within the error of the absorbance measurements.) On examination, however, the 2:1 films were shown to have degraded rather than leached, most likely due to their unstable nature at the time of measurement. This instability was discussed in the last section and is related to low water content. The R=4 slide was shown to be very stable. Fig. 6.14 shows that the absorbance remained the same over a period of 900 h in static water.

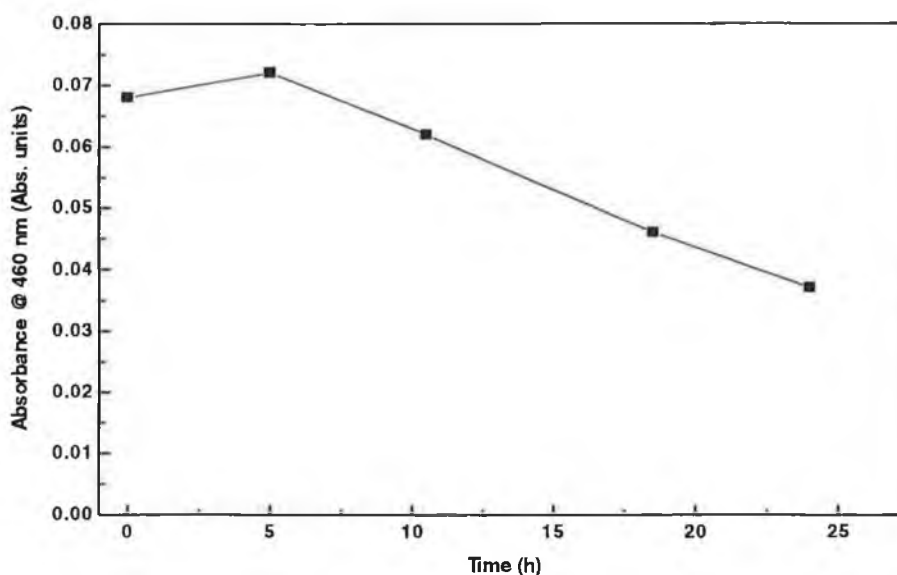


Figure 6.13: Absorbance of unstabilised R=2 TEOS film in water as a function of time.

Slides were also monitored in stirred water. The same procedure as above was carried out using stabilised R=2 slides in agitated (stirred) water, and the absorbance reading at 460 nm remained unchanged throughout. This can be seen in Fig 6.15. As mentioned in Section 3.7, due to the fact that the ruthenium complex is added in at the start of the fabrication procedure, the hydrolysis and condensation reactions take place in the presence of the dye complex and the matrix builds up around it. As a consequence, the complex is fully entrapped, and tightly held within the structure, and hence no leaching of dye occurs.

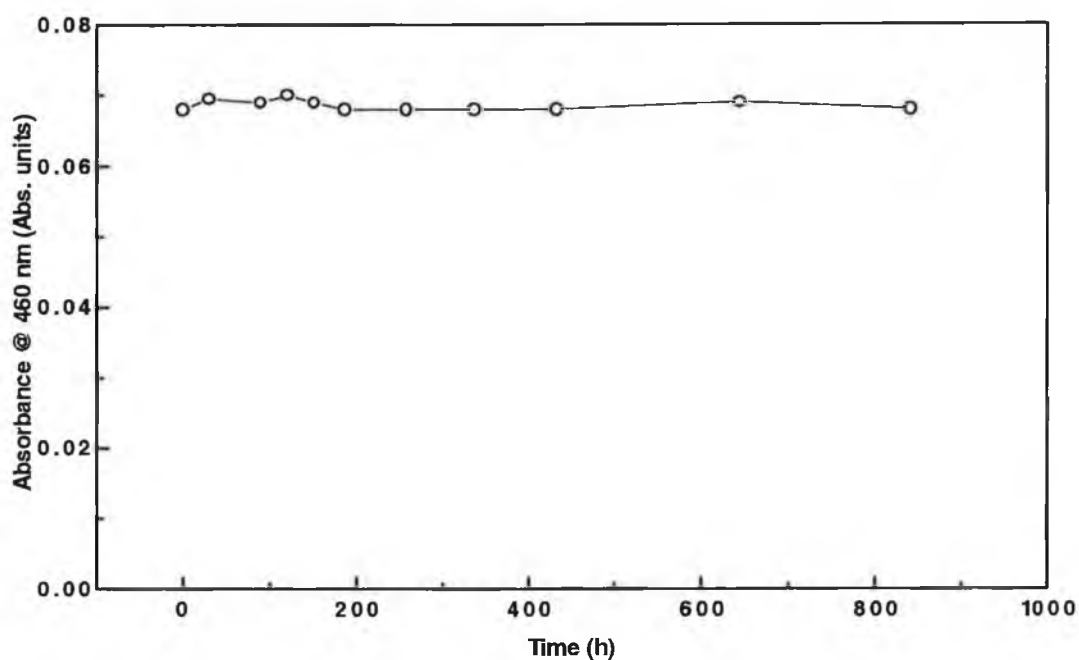


Figure 6.14: Absorbance of R=4 stabilised film in water as a function of time.

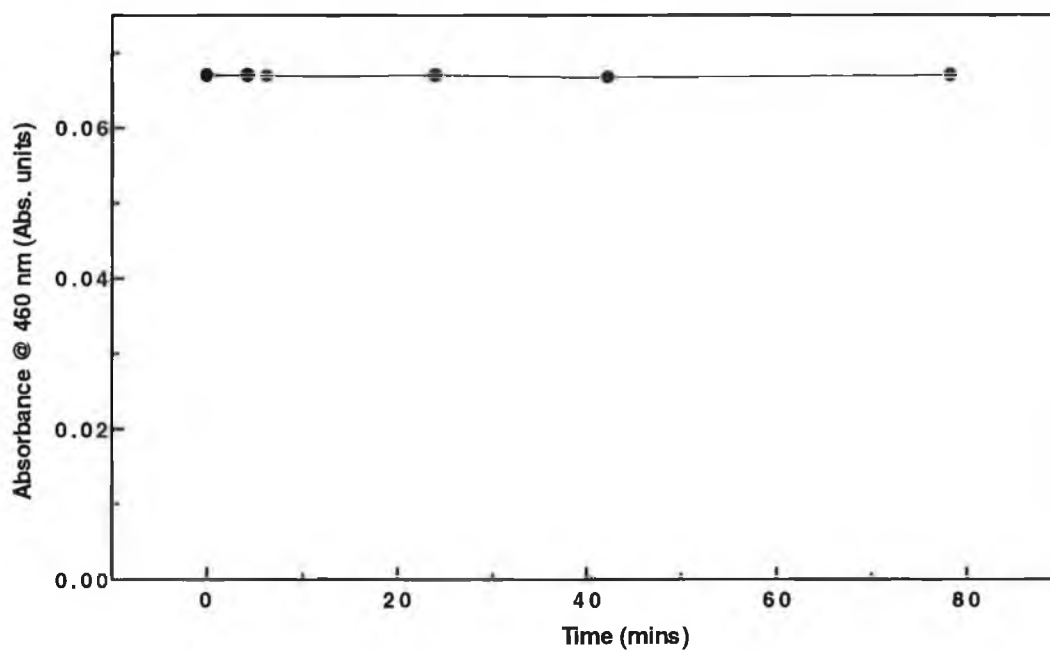


Figure 6.15: Absorbance of R=2 stabilised film in agitated (stirred) water as a function of time.

6.3 Organically modified films

Films fabricated using a silicon alkoxide precursor are hydrophilic with a high surface coverage of silanol groups.^{1,8} The main reasons for the interest in surface silanol groups is because surface coverage largely determines the adsorption behaviour and consequently the surface reactivity.³ In this case, as was briefly discussed in Section 6.2.2.1, the presence of silanol groups on the surface (chemisorbed water), encourages the adsorption of surface water (physisorbed water). It is thought that the physisorbed water may reduce the effective pore size preventing total quenching. This, coupled to the smaller gas partitioning effect at the hydrophilic surface leads to the lower quenching response. As was mentioned earlier, for a viable DO sensor a high overall quenching response is necessary to attain good resolution at low oxygen concentrations. By analogy with the Clark electrode and polymer-based sensors, it was thought that increasing the hydrophobicity of the surface would increase the quenching response in water, by excluding the water and causing oxygen gas to partition out of solution and pass into the pores as mentioned in Section 6.2.2.1. A number of methods were investigated, for example, silylation of the surface, coating with a silicone rubber barrier layer, and organic modification of the sol-gel structure.

Silica can be made hydrophobic by treatment with derivatisation agents such as dimethyldichlorosilane (DMDCS).⁹ Silylation involves post-treating the surface with a silylating agent such as DMDCS, whereby the silicon along with the two methyl groups bonds to the surface, liberating HCl in the process. This was tried and was not found to improve the quenching response in DO, which was probably due to the reduction in size of the pores due to the large size of the bonding group.

The second method involved coating TEOS films with a silicone rubber film or a Teflon film in an attempt to confer some degree of hydrophobicity on the sensing layer. Silicone rubber is known for its high solubility and permeability to oxygen, as is Teflon. Again in this case, no appreciable increase in DO quenching resulted, which may be due to the permeability of the barrier layer to humid gas, and to the hydrophilicity of the underlying TEOS film. This will be discussed in the next chapter, but it is thought that because the barrier layer is a hydrophobic coating, partitioning into the gas phase occurs. This oxygen gas is highly humid, and passes through the oxygen permeable layer into the pores of the sensing layer. When the humid gas hits the largely hydrophilic TEOS surface, it condenses forming a water barrier layer (physisorbed water), which reduces

quenching of the ruthenium complex.

Successful enhancement of the surface hydrophobicity was achieved, however, by organic modification of the sol-gel films. This requires addition of an organically modified precursor along with, or in place of, the silicon alkoxide precursor in the fabrication procedure.

Organically modified precursors are similar to their silicon alkoxide counterparts in having a central silicon metal ion surrounded by alkoxy (-RO) groups, but having an organic group substituted for one of the alkoxy groups. For example, methyltriethoxysilane, MTEOS, has a methyl group in place of the fourth ethoxy group i.e. $\text{CH}_3\text{Si}(\text{OC}_2\text{H}_5)_3$. In organically modified films, the organic groups partially replace the hydroxyl groups. Due to the poor affinity of organic groups for water, the surface hydrophobicity is increased.

The following involves a comprehensive study of organic modification of sol-gel films, and its effects on the quenching by dissolved oxygen of the complex. Oxygen has a relatively low solubility in water whereby the concentration of oxygen in water in equilibrium with air is only 9.2 ppm at 20°C and standard atmospheric pressure. Increasing the surface hydrophobicity and thereby increasing the quenching response in water compensates to a certain extent for the low DO solubility. This increasing hydrophobicity is demonstrated by Fourier Transform Infrared Spectroscopy measurements shown in Section 6.3.2, which show an increase in the intensity of the Si-R bands coupled with a decrease in the water band, with modified precursor content. Tailoring in aqueous phase is also possible by varying the R value of modified sol-gel films. This is discussed in Section 6.3.3. Thickness stabilisation measurements are detailed in Section 6.3.4. Finally, parallels are drawn between gas-phase and aqueous phase quenching responses for modified films. Quenching responses in the two phases reflect opposing trends and this will be dealt with in Section 6.3.5.

6.3.1 Aqueous phase quenching behaviour

Preliminary investigations into the effect of modified precursors on the quenching response of the films in aqueous phase involved the use of triethoxysilane, HTEOS, the first member of the organically modified TEOS-based group. A sol with R value of 2 was fabricated as documented in Section 4.2, except that a 1:1 mixture of HTEOS:TEOS replaced the TEOS. The films were dip-coated and dried

as normal. The quenching response in aqueous phase of the R=2 HTEOS:TEOS 1:1 films can be seen in Fig. 6.16, where Q_{DO} was found to be 38%. This is almost twice that of the corresponding TEOS films, and it was concluded at this stage that the likely source of this enhancement was the increased hydrophobicity of the surface which increases the overall quenching response in dissolved oxygen. R=4 HTEOS:TEOS 1:1 films were also fabricated and their Q_{DO} was found to be 30%, lower than the R=2 film, which corresponds to the results obtained for their TEOS counterparts. However, as these films were monitored periodically to investigate their stability with time, it was found that they degraded irreversibly.

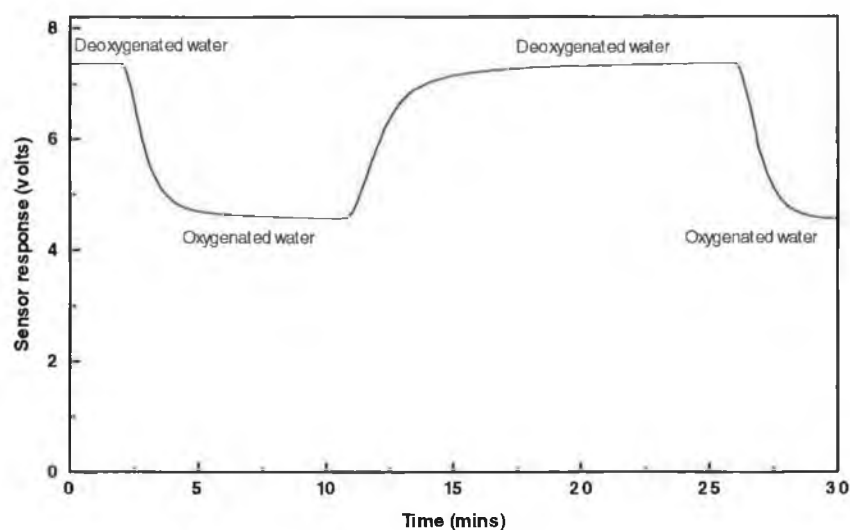


Figure 6.16: Quenching response of R=2 HTEOS:TEOS 1:1 films in aqueous phase

The quenching response of the R=2 and R=4 HTEOS:TEOS 1:1 films reverted to that of the equivalent TEOS films [$Q_{DO}(R=2) = 20\%$ and $Q_{DO}(R=4) = 6\%$] over a period of two weeks. It is thought that because the organic part of the precursor is a hydrogen atom, it is easily hydrolysed to form Si-OH groups on the surface, eventually yielding effectively a TEOS film. To continue this investigation, coating sols were fabricated using increasing amounts of methyltriethoxysilane, MTEOS, to TEOS, ranging from 100% TEOS to 100% MTEOS. Coating sols were fabricated as described previously, but were not aged in the oven after stirring due to their short gelation time, t_g . Films were coated directly after stirring and then dried at 70°C for 18 h. The quenching response to dissolved oxygen was measured for each of the films, and this data can be seen in Table 6.2.

SOL-GEL FILM	Q_{DO} ($\pm 2\%$)%
R=2 TEOS	20
R=2 MTEOS:TEOS 1:1	56
R=2 MTEOS:TEOS 2:1	62
R=2 MTEOS:TEOS 3:1	70
R=2 MTEOS	73

Table 6.2: Q_{DO} of TEOS and of modified MTEOS/TEOS films.

It will be shown in Section 6.3.2 that the number of surface SiCH_3 groups increases, as expected, with increasing ratio of MTEOS to TEOS.¹⁰ The SiCH_3 groups replace the SiOH groups, thereby enhancing the hydrophobicity, as the CH_3 groups have a lower affinity for water. The changes in Q_{DO} shown in Table 6.2 may therefore be explained by an increased partitioning of oxygen into the gas phase, resulting from an increase in surface hydrophobicity. Fig. 6.17 shows the quenching response of R=2 MTEOS:TEOS 3:1 films in aqueous phase.

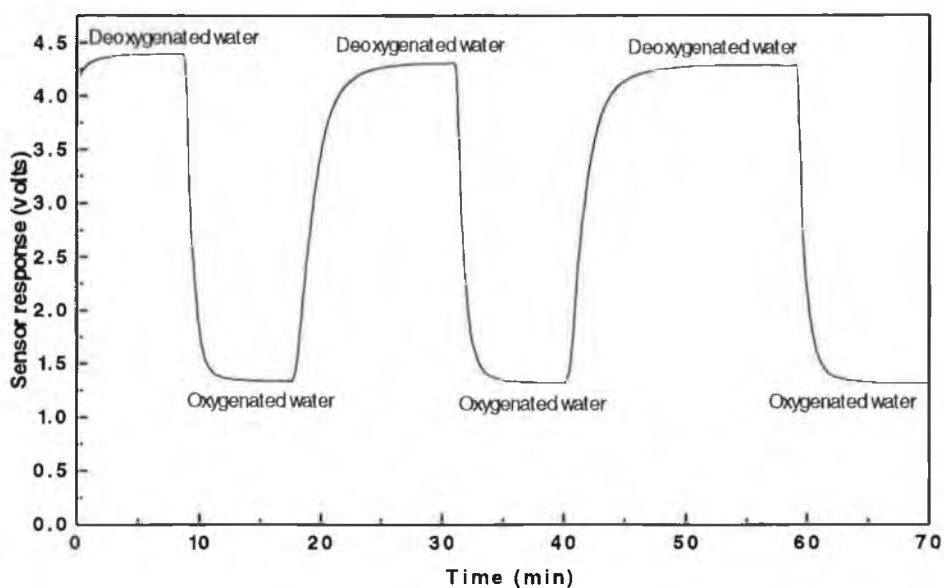


Figure 6.17: Quenching response of R=2 MTEOS:TEOS 3:1 film in aqueous phase.

The value of Q_{DO} for the R=2 MTEOS:TEOS 3:1 film is 70% as shown in Table 6.2. Although the response time appears to be much longer than in the gas

phase, this is mainly due to instrumental considerations such as the volume of the sample cell and the DO mixing / delivery system. The sensing film exhibits good reversibility, excellent repeatability and a very high signal-to-noise ratio.

Figure 6.18 shows Stern-Volmer plots for the R=2 MTEOS films. For all these plots there is a larger linear region at concentration > 5 ppm oxygen. The curvature at low oxygen concentrations was observed by many authors¹¹ and is not fully explained, but may be due to site heterogeneity in the sol-gel matrix. The K_{SV} values tabulated in Table 6.3 are calculated from the linear part of the plots. The 100% MTEOS film has the largest K_{SV} value which, as K_{SV} increases with Q_{DO} , is consistent with the Q_{DO} data shown in Table 6.2.

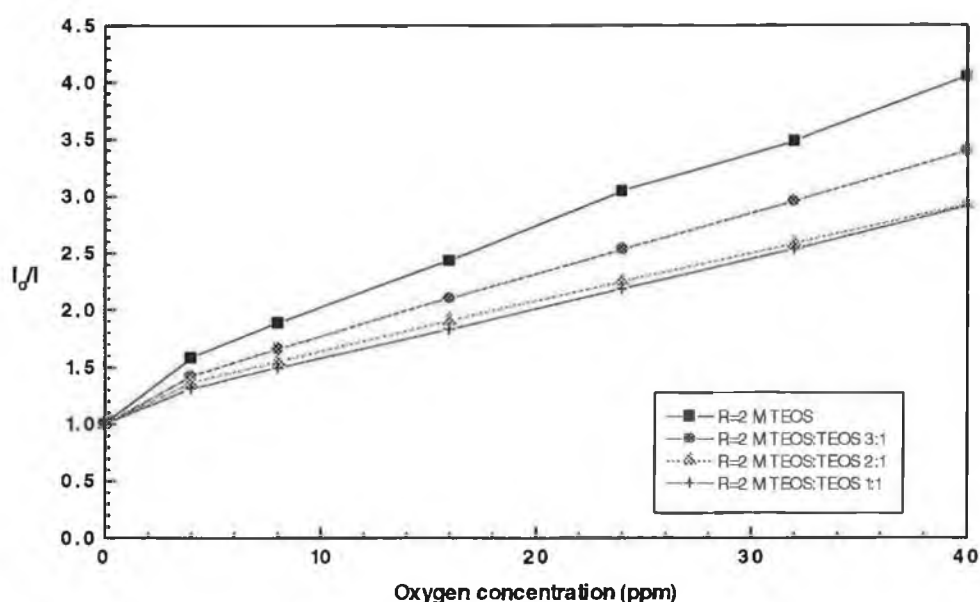


Figure 6.18: Stern-Volmer plots for R=2 MTEOS films.

FILM	STERN-VOLMER CONSTANT, K_{SV}
R=2 MTEOS	2220 M^{-1}
R=2 MTEOS:TEOS 3:1	1800 M^{-1}
R=2 MTEOS:TEOS 2:1	1440 M^{-1}
R=2 MTEOS:TEOS 1:1	1266 M^{-1}

Table 6.3: Stern-Volmer constants, K_{SV} for R=2 MTEOS films.

As was seen in Chapter 2, the Stern-Volmer constant, K_{SV} , is proportional to αD where α is the oxygen solubility in the matrix, and D is the diffusion coefficient of oxygen in the film. The predominant factor governing the quenching behaviour of these films in DO is hydrophobicity. It is proposed that the observed increase in DO quenching and the accompanying increase in Stern-Volmer constant as a function of hydrophobicity can be correlated with an increase in α , the solubility of oxygen in the sol-gel film in aqueous phase. This is in contrast to the argument in Section 6.2.2.1 for TEOS gas phase sensing, where α was assumed to be constant and changes in film microstructure resulted in changes in D . It is likely that in DO quenching, some of the oxygen is transported purely in aqueous phase as opposed to being partitioned out at the surface, as mentioned in the case of TEOS films in Section 6.2.2.1. However, it is assumed that the predominant effect for modified films is partitioning due to the hydrophobicity of the surface.

This investigation was further extended to include the third member of the aliphatic group, ethyltriethoxysilane, ETEOS. Two ETEOS-based films were fabricated, R=2 ETEOS:TEOS 1:1 and R=2 ETEOS. Again, due to the short gelation time of ETEOS sols, no aging was carried out, and the films were dip-coated immediately after stirring. Having measured the overall quenching response of the ETEOS films, it was found that extending the aliphatic group of the organically modified precursors increases the overall quenching response of the films in water. Q_{DO} for R=2 ETEOS:TEOS 1:1 films was found to be 73% and Q_{DO} of R=2 ETEOS films was 80%. Instead of having Si-CH₃ groups on the surface, as for MTEOS films, the longer Si-C₂H₅ groups provide surface coverage. Thus by extending the aliphatic group of the organically modified precursors to incorporate in this case, ETEOS, it is proposed that the hydrophobicity increases as does the solubility of oxygen in the film, hence a larger value of Q_{DO} .

Up to this, the organically modified precursors used had aliphatic organic groups, for example, methyl and ethyl. Phenyltriethoxysilane, PhTEOS, has the same basic structure as the previous modified precursors, but has an aromatic or ring organic group rather than an aliphatic group, whose structure is C₆H₅. It was thought that PhTEOS would increase the quenching response yet further. Films were fabricated, dip-coated and dried as before. Fig. 6.19 shows the quenching response of R=2 PhTEOS films in aqueous phase. Q_{DO} was found to be only 61% for these PhTEOS films. This is intermediate between R=2 MTEOS:TEOS 1:1 and R=2 MTEOS:TEOS 2:1 films. The reason for this low quenching response in fully

oxygenated water is thought to be due to steric hindrance caused by the bulky nature of the phenyl groups. This result corroborated by Schmidt's work where it was found that phenyl groups can cause steric hindrance.¹²

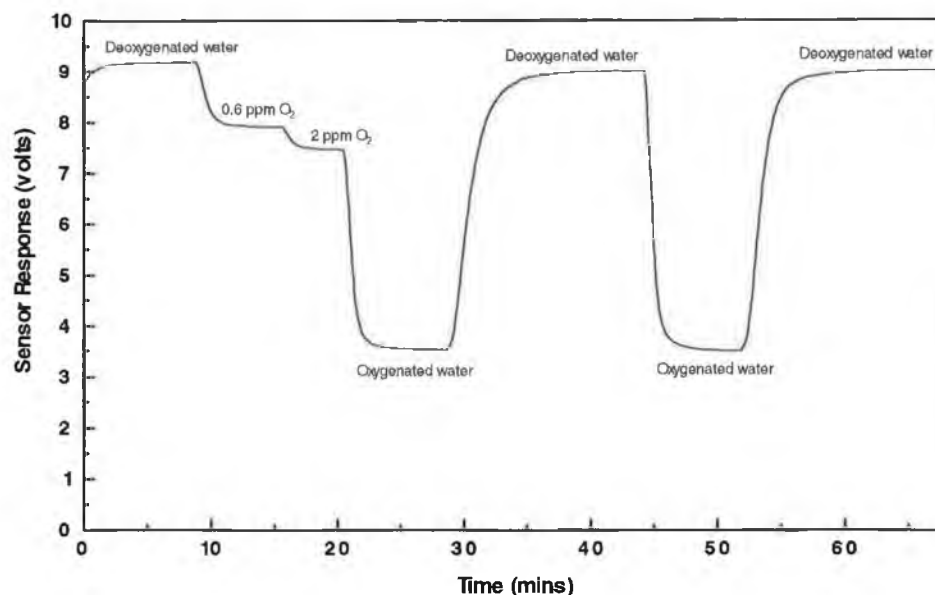


Figure 6.19: Quenching response of R=2 PhTEOS film in aqueous phase.

From this study of the quenching response of various modified precursors, it was found that by both increasing the ratio of modified precursor to silicon alkoxide precursor, and by extending the aliphatic group of the modified precursor, the overall quenching response of the films in aqueous phase increased. This is as a result of alkyl groups, which have a poor affinity for water, being substituted for hydroxyl groups on the film, thereby increasing the hydrophobicity of the film which we propose is related to the solubility of oxygen in the film. This increased hydrophobicity increases the degree of gas partitioning at the film / water interface causing an increase in Q_{DO} . The results of the following section establish that increased hydrophobicity is correlated with an increased concentration of modified precursors and longer organic aliphatic groups.

6.3.2 Fourier Transform Spectroscopy

Fourier Transform Infrared (FTIR) Spectroscopy has been used by a number of groups to probe the microstructure of silica.^{10, 13} Innocenzi et al,¹⁰ in particular, used FTIR spectroscopy to investigate the composition of MTEOS films, and compare it to that of TEOS films. This work indicates that the molecular water band @ 3400 cm^{-1} and the Si-OH band @ $910\text{-}940\text{ cm}^{-1}$ decrease with increasing MTEOS content, whereas the Si-CH₃ bands @ 1260 cm^{-1} and $2900\text{-}3000\text{ cm}^{-1}$ increase.

This technique was used in this laboratory to investigate the composition of the organically modified films.¹⁴ Silicon wafers were coated with various modified films, and infrared spectra, in the range, $400\text{ to }4000\text{ cm}^{-1}$ were obtained by FTIR spectroscopy. In order to get a consistent idea of how the structure is changing for each film, the ratio of Si-O-Si peaks to Si-OH peaks was evaluated, and the intensity of the molecular water band and the C-H band @ 3000 cm^{-1} was recorded and can be seen in Table 6.4 below. It was possible to use the values at the peaks as the band widths did not vary much for each film. The overall quenching response, Q_{DO} , of the modified films in water is also included and is consistent with the increased hydrophobicity of ETEOS and MTEOS films relative to mixed MTEOS/TEOS films.

Bands	Organically modified sol-gel films		
	ETEOS	MTEOS	MTEOS:TEOS 1:1
Si-O-Si/Si-OH	1.76	1.72	1.46
C-H @ $\sim 3000\text{ cm}^{-1}$	0.1	0.07	0.03
Water @ 3400 cm^{-1}	Negligible	0.9	0.8
Quenching response	80%	72%	54%

Table 6.4: Data obtained from FTIR spectroscopy of modified sol-gel films

As can be seen, the results are as expected apart from the water band in the MTEOS film which is slightly greater than that of the MTEOS:TEOS 1:1 film. As the ratio of modified precursor to TEOS increases and the aliphatic group is

extended, the ratio of Si-O-Si to Si-OH increases as does the C-H band at 3000 cm^{-1} , confirming the increasing hydrophobicity of the films, and thus corroborating the results from Section 6.3.1.

6.3.3 Tailoring of quenching response

There is an inherent non-linearity in the Stern-Volmer equation (Eqn. 2.12), which can be seen more clearly if the equation is differentiated. Equation 6.3 shows the differentiated version of the Stern-Volmer equation

$$\frac{dI}{d(pO_2)} = \frac{-K_{SV} I_0}{(1+K_{SV} pO_2)^2} \quad (6.3)$$

and all the terms are as were defined previously in Section 2.3. It is evident from this equation that the rate of change of intensity decreases with increasing partial pressure of oxygen. Referring to Figures 6.2, 6.3 and 6.4, it is clear that there is a greater sensitivity to low oxygen concentrations than higher concentrations, regardless of the R value, in gas phase. The degree of this sensitivity, however, does depend mainly on the porosity of the sample, which affects the diffusion of gas through the sample. As seen previously for TEOS films in gas phase, as the overall quenching response increases, so too does the quenching response in low oxygen concentrations. This results in a higher value of K_{SV} , the Stern-Volmer constant.

By analogy with the R-value tailoring of the quenching response of TEOS films in gas phase, quenching in MTEOS films can also be tailored for both gas and aqueous phase measurements. Figure 6.20 shows fluorescence intensity data as a function of dissolved oxygen concentration for R=2 and R=4 MTEOS films.

The behaviour is similar to that of TEOS in gas phase whereby the lower R value film has a larger quenching response and a higher sensitivity at low oxygen concentrations. The quenching response in 4 ppm of DO is 38% for R=2 MTEOS films, but only 20% for R=4 MTEOS films. The calibration data is more linear for R=4 MTEOS films, which is consistent with results for TEOS films in gas phase.

As for TEOS films, it is proposed that this behaviour is consistent with larger average pore size, and hence a larger diffusion coefficient for R=2 films compared with R=4 films. This demonstrates how the microstructure of modified films can be

tailored to optimise the quenching response in a particular dissolved oxygen concentration range.

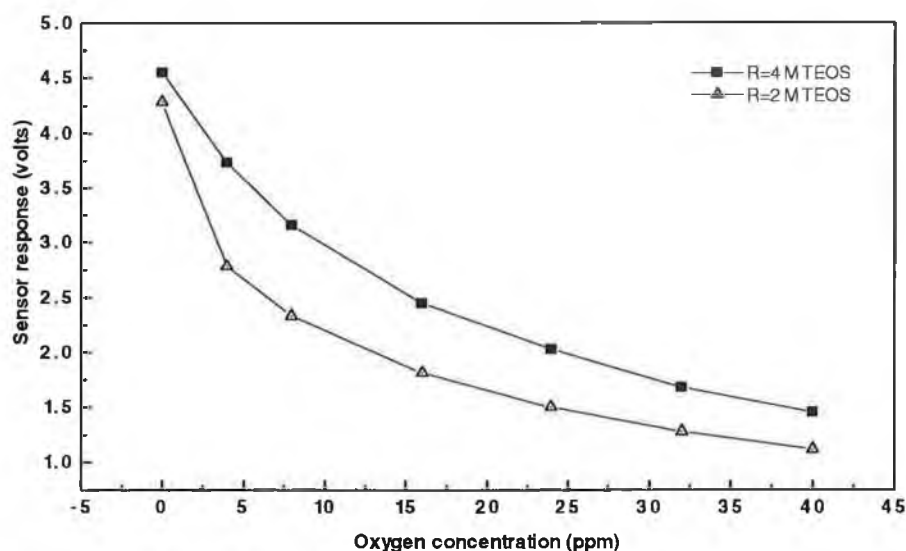


Figure 6.20: Calibration data for R=2 and R=4 MTEOS films in DO.

6.3.4 Thickness stabilisation

Section 6.2.3 dealt with the thickness stabilisation of the microstructure of TEOS-based films. These data showed that, the time it took for the film properties to reach a steady state condition such that film thickness and porosity do not change, is significantly different for R=2 and R=4 TEOS films. For R=2 films, the film thickness decreased gradually over approximately 80 days, while for R=4 films, their thickness stabilised in approximately 20 days. In this work, two sets of modified films have been monitored with regard to the stabilisation of the thickness of the films as a function of time. Modified MTEOS:TEOS 1:1 films with R values of 2 and 4 were fabricated and their thickness was monitored as a function of time after drying. The results are shown in Fig. 6.21.

This is a similar result to that shown in Fig. 6.12. Films with R=4 MTEOS:TEOS 1:1 take only days for their thickness to stabilise. Similar results were found for R=4 MTEOS and R=4 MTEOS:TEOS 3:1 films but these data are not shown here. The maximum stabilisation time is 10 days for R=4 MTEOS modified films. R=2 films have a much slower stabilisation time, which is analogous with TEOS-based films. However, because the gelation time is faster for modified films than for TEOS-based films, it can be assumed that the stabilisation time of the modified

films will be faster also. Fig. 6.21 shows inconclusive data for R=2 thickness stabilisation, however in comparison to Fig. 6.12 for TEOS films, the percentage change of thickness with time is much smaller in this case.

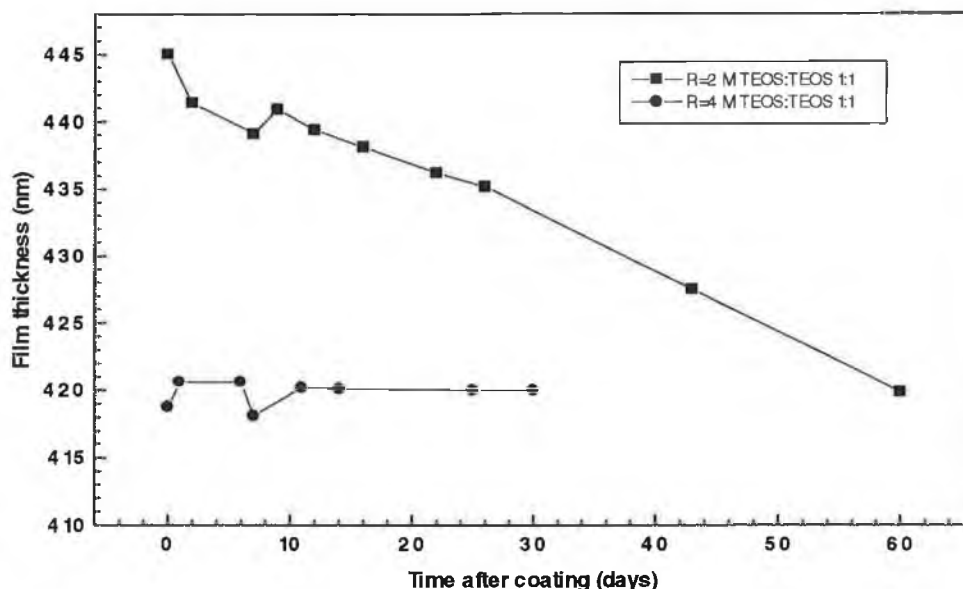


Figure 6.21: Thickness stabilisation of R=2 and R=4 MTEOS:TEOS 1:1 films.¹⁴

6.3.5 Gas phase measurements

Quenching response measurements in gas phase for modified films were carried out in order to extend the investigation of modified films. A comparison between gas phase and aqueous phase quenching response measurements can give an indication as to the structure of the films, and the mechanisms by which quenching occurs.

Gas phase quenching measurements were carried out using planar substrates coated with modified films. The characterisation system used was that seen in Fig. 5.6, and the gas mixing system as shown in Fig. 5.3. Table 6.5 shows the quenching response in both gas and aqueous phase for modified films with varying TEOS/MTEOS and TEOS/ETEOS ratios. The Q_{DO} values are shown for comparison purposes.

Clearly, Q in gas phase decreases as a function of MTEOS content, while Q in dissolved oxygen increases with MTEOS content. The quenching behaviour in DO, as seen in Section 6.3.1, can be correlated with the surface hydrophobicity,

whereby the increase in Q_{DO} with MTEOS content results from increasing hydrophobicity due to the increasing number of CH_3 groups on the film surface. This in turn increases the partitioning effect, whereby more gas comes out of solution and the solubility of the gas in the film is increased.

	Q_{gas}	Q_{DO}
MTEOS/TEOS 1:1	93%	56%
MTEOS/TEOS 2:1	92%	62%
MTEOS/TEOS 3:1	89%	70%
MTEOS	85%	73%
ETEOS/TEOS 1:1	92%	72%
ETEOS	87%	80%

Table 6.5: Overall quenching response in gas and aqueous phase for R=2 organically modified sol-gel films.

In gas phase, on the other hand, if hydrophobicity effects are assumed to be minimal, the decrease in Q_{gas} with MTEOS content, and in going from MTEOS films to ETEOS films, indicates a decrease in diffusion coefficient, D , due to a partial blocking of the pores by the CH_3 groups, and in the case of ETEOS, by the even larger ethyl (C_2H_5) groups.

From the above discussion, it is evident that the quenching data in Table 6.5 reflect two opposing trends, in the case of gas phase and aqueous phase quenching respectively. In gas phase, it is proposed that the effect of increasing MTEOS content results in decreased diffusion coefficients due to smaller average pore size, hence causing a reduction in the quenching coefficient, Q_G . It is reasonable to assume that the solubility effect on the ormosil films in gas phase is much smaller than the diffusion effect. In DO, however, hydrophobicity is the predominant effect and it is proposed that the increase in quenching with increasing hydrophobicity is related to increasing solubility of oxygen in the sol-gel matrix.

6.4 Conclusions

In this chapter, an optimum sensing film has been identified for optical dissolved oxygen sensing. Although TEOS films were found to be efficient gas phase sensors, due to the predominantly hydrophilic nature of the surface, the quenching response in water was quite low. This in turn led to poor resolution in lower DO concentrations. It was necessary to increase the quenching response in water to develop a viable DO sensor. Modifying the TEOS surface using non-invasive techniques like silylation and coating with silicone rubber or Teflon did not change the quenching response. Fabrication of films using organically modified precursors resulted in the substitution of hydrophilic hydroxyl groups with alkyl groups which have a poor affinity for water, thus making the surface more hydrophobic. Increasing the MTEOS content, and extending the aliphatic group from HTEOS to MTEOS to ETEOS increased the hydrophobicity, making the partitioning effect more efficient. This in turn increased the quenching response in water. R=2 MTEOS and R=2 ETEOS have the highest Q_{DO} values and have been found to be the optimum films for DO sensing. The performance of these films as sensors with regard to their properties such as response time, limit of detection, and stability of quenching response, as well as the effect of temperature and interferent species on the sensors will be investigated in Chapter 7.

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CHAPTER 7: PERFORMANCE OF OPTIMAL SENSOR

7.1 Introduction

Of the films studied so far in this work, the optimum preparations for optical dissolved oxygen sensing were identified in the previous chapter. R=2 MTEOS and R=2 ETEOS films were chosen primarily because of their high quenching response to dissolved oxygen, but also due to their high quenching stability, good resolution at low DO concentrations and their excellent reversibility when cycling through oxygenated and deoxygenated water. As seen in Table 6.5, the overall quenching response in DO of R=2 ETEOS films is 80%, and that of R=2 MTEOS films is 73%. Experiments were carried out on the optimum films to test their performance under certain conditions. The results of these experiments are documented in this chapter.

7.2 Quenching response of optimal films in aqueous phase

Films fabricated with MTEOS have a high surface coverage of methyl ($-\text{CH}_3$) groups, and this, as was seen in Chapter 6, confers a high degree of hydrophobicity on the films. There is a large partitioning effect increasing the overall quenching response. The surface of ETEOS films is largely covered with ethyl ($-\text{C}_2\text{H}_5$) groups, and it appears that these groups further increase the partitioning effect resulting in a greater overall quenching response of 80% in dissolved oxygen.

An important property of a practical DO sensor in some applications is good resolution at low oxygen concentrations. The high sensitivity achieved with the optimised films is further enhanced at low oxygen concentrations as discussed in Section 6.3.3. This results in a higher resolution at low concentrations and a lower LOD.

Figures 7.1 and 7.2 show the calibration data for R=2 MTEOS and R=2 ETEOS films. The large sensitivity at low oxygen concentrations is evident. For R=2 ETEOS films, the quenching response in 0.6 ppm O_2 is 35%, which indicates the suitability of these films for sensing in very low (ppb) oxygen concentrations (see Section 7.6 on sensor resolution and limit of detection).

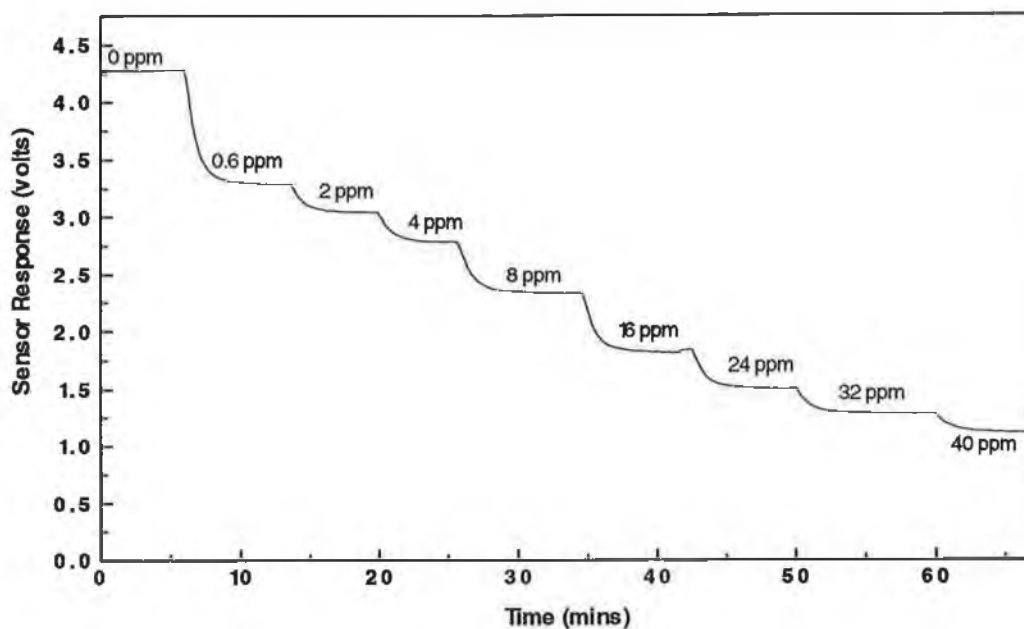


Figure 7.1: Calibration data for R=2 MTEOS film

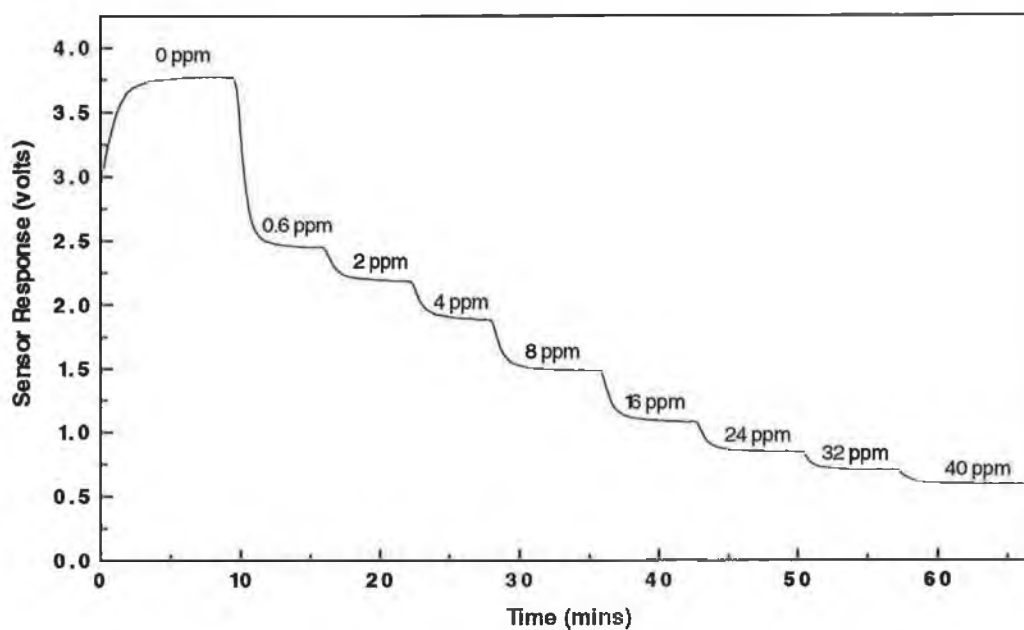


Figure 7.2: Calibration data for R=2 ETEOS film.

The quenching response of R=2 MTEOS films in 0.6 ppm is 23%, which also indicates good sensor resolution at low concentrations. A comparison of Stern-Volmer plots for the optimum films can be seen in Fig. 7.3.

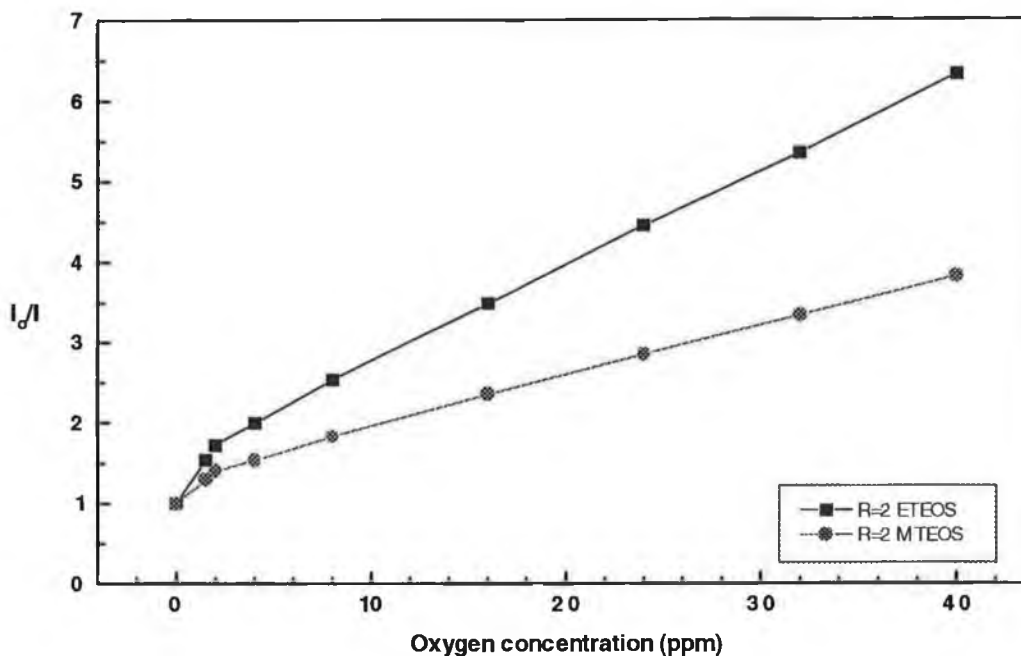


Figure 7.3: Stern-Volmer plot of R=2 MTEOS and R=2 ETEOS films in DO.

It is clear from this comparison that the ETEOS film is more sensitive than the MTEOS film. The Stern-Volmer constant, K_{SV} , which was calculated from the linear part of the graph above 5 ppm oxygen concentration, was found to be 3950 M^{-1} for the ETEOS films, and 2220 M^{-1} for the MTEOS films. However, as seen in Table 6.3, both of these films have K_{SV} values considerably larger than those of the MTEOS/TEOS mixtures.

7.3 Sensor response time

The inherent response time of the sensing film is difficult to measure precisely. We define response time here as the time it takes for the intensity in totally deoxygenated water to change to that in fully oxygenated water. Effectively, it is the turn-on time of the sensor. The turn-off time is the reverse situation, and is known here as the recovery time. Using the normal characterisation cell, it was not possible to deduce the sensor response time. This was due to the fill time of the relatively large volume cell (approximately 230 s), and the delivery time of the calibrated solutions to the sensing substrate. The procedure for delivering

calibrated oxygenated and deoxygenated solutions to the cell involved first oxygenating and deoxygenating for approximately 15 minutes in a large reservoir as shown in Fig. 5.9. The gas was then switched off and water circulation and data acquisition started simultaneously. For the large 23 mL cell, the fill time was calculated to be approximately 230 s, while the fill time of the smallest (1 mL) cell was approximately 10 seconds. This fill time, the oxygen diffusion coefficient through the flowing water and the diffusion rate associated with partitioning of oxygen into the film, all contribute to the response time. The diffusion coefficient of oxygen in water at infinite dilution and at room temperature is $2 \times 10^{-9} \text{ m}^2/\text{s}$.¹ The diffusion coefficient of oxygen in a sol-gel film is typically $2 \times 10^{-7} \text{ m}^2/\text{s}$.² In this work, it has not been possible to extract individual diffusion coefficients from the quenching data. However, this whole area is still under investigation.

Fig. 7.4 shows the quenching response of a R=2 MTEOS film to dissolved oxygen using the 1 mL glass cell, and Fig. 7.5 shows that of the R=2 ETEOS film.

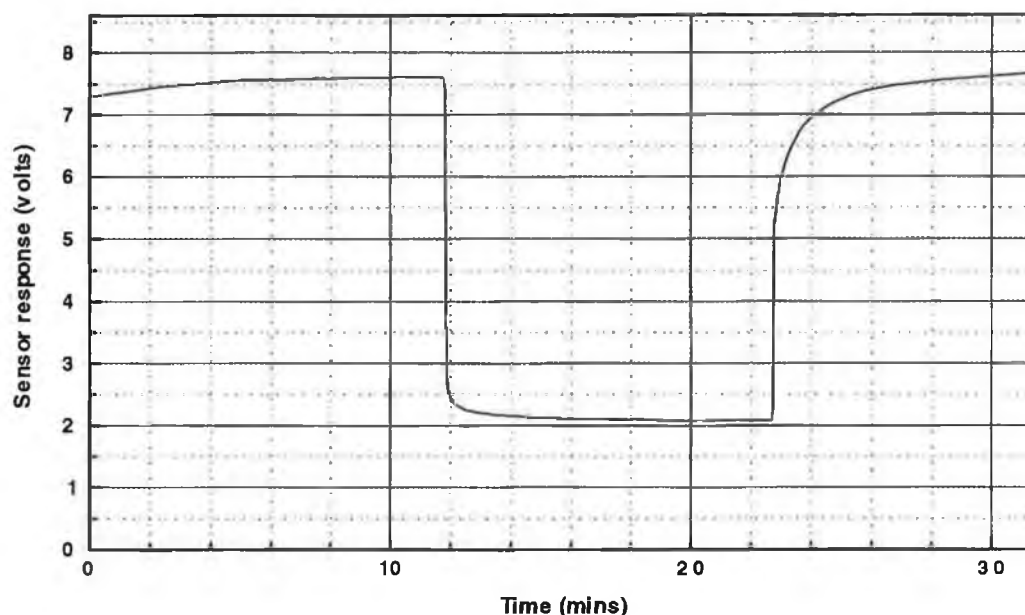


Figure 7.4: Quenching response of R=2 MTEOS films in DO using the 1 mL cell, indicating the fast response time of the sensor.

From these graphs, the short response time of the sensor is evident. An obvious feature of the quenching response is the difference between the response time and the recovery time. The latter is considerably longer than the former. This

issue is treated in detail by Mills *et al.*, and has been modelled as predicted by the Stern-Volmer relation.³ These scans were performed a number of times to estimate the response times of the films. The normal way of describing sensor response times is in terms of t_{90} , which is the time it takes for the signal to drop to 90% of the full-scale value for response time, and the time it takes to rise to 90% of the full-scale value for recovery time.

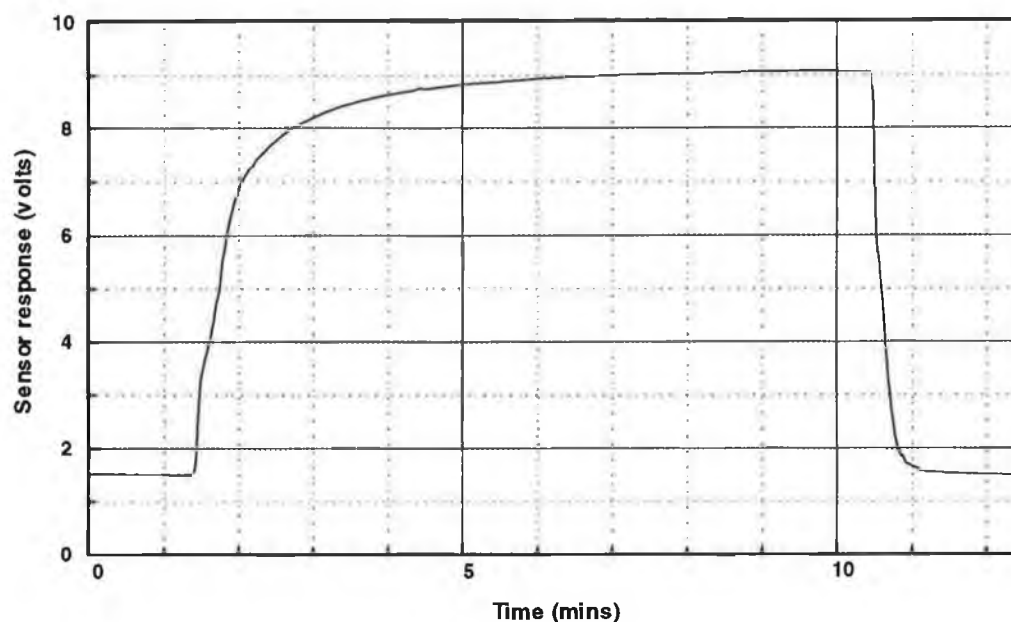


Figure 7.5: Quenching response of R=2 ETEOS film in DO using the 1 mL cell for sensor response time estimations.

The value of t_{90} was averaged over three estimations with a standard deviation of 3 seconds, for both the response time and the recovery time of R=2 MTEOS and R=2 ETEOS films, and these data are tabulated in Table 7.1.

	Response time ± 3 (sec)	Recovery time ± 3 (sec)
R=2 MTEOS	9	125
R=2 ETEOS	17	123

Table 7.1: Estimates of response and recovery times for optimum films using 1 mL cell.

There is little difference between the recovery times for both films, but the response time for the R=2 MTEOS film is shorter than that of the R=2 ETEOS film, possibly due to lower steric hindrance of the methyl groups compared with the larger ethyl groups.

7.4 Humidity tests

Humidity tests were performed on the optimum films to test their response to humidified gas. Given the principle of operation of the sensor detailed earlier, it was expected that 100% humidified oxygen gas would give the same response as fully oxygenated water. If the response in humidified gas and in aqueous phase were the same, then calibration of the sensor could be performed more readily in humidified gas rather than in liquid, and this would make the process easier and faster.

Gas was humidified using the system shown in Fig. 5.10(ii). Cycling the gas through the four gas washing bottles forced it to adsorb water and become highly humid. A humidity sensor was incorporated into the set-up which measured the relative humidity (RH) of the gas. This was found to be 99.8% RH for all the experiments. Quenching response, Q , of all ormosil films was measured in humidified gas, $Q_{\text{gas (humid)}}$, the results of which are shown in Table 7.2. Quenching response data in gas phase ($Q_{\text{gas (dry)}}$) and DO are shown for comparison purposes.

	$Q_{\text{gas (dry)}}$	$Q_{\text{gas (humid)}}$	Q_{DO}
R=2 ETEOS	87%	85%	80%
R=2 ETEOS:TEOS 1:1	92%	80%	73%
R=2 MTEOS	85%	77%	74%
R=2 MTEOS:TEOS 3:1	89%	75%	71%
R=2 MTEOS:TEOS 2:1	92%	71%	62%

Table 7.2: Overall quenching response in gas phase, humid gas and aqueous phase for modified films.

Figures 7.6 and 7.7 show the quenching response of R=2 MTEOS films in dry gas and in humid gas.

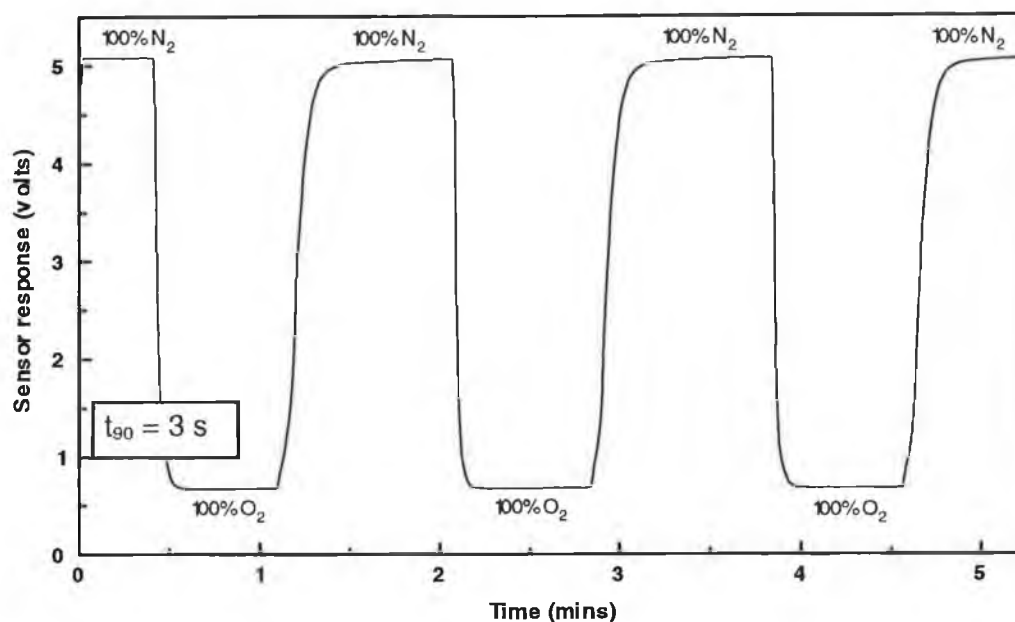


Figure 7.6: Quenching response of R=2 MTEOS film in gas phase.

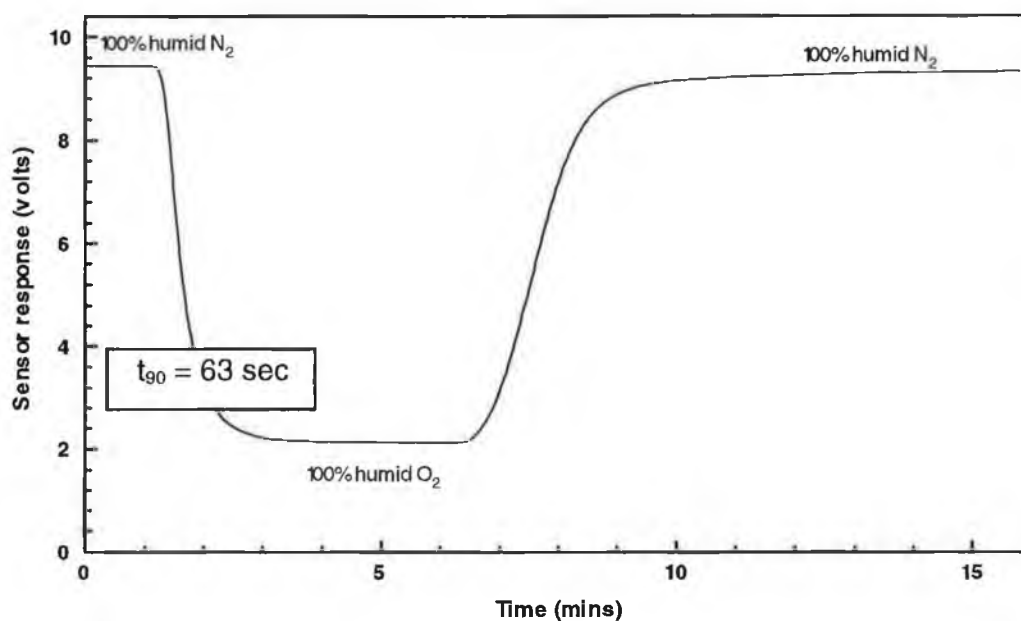


Figure 7.7: Quenching response of R=2 MTEOS films in humidified gas (t_{90} indicates response time).

The t_{90} detailed is the response time of the film in both cases. The films with the greatest degree of hydrophobicity, R=2 ETEOS and R=2 MTEOS, have Q values

in humid gas and water which are most similar. The lower the film hydrophobicity, the greater the difference in quenching response between the three phases. This difference is likely to be due the physisorbed water on the more hydrophilic surface causing a reduction in Q_{DO} and Q_{humid} as discussed in Section 6.3 for the case of Q_{DO} . These are complex issues involved in interpreting the humidity data, and in comparing it with the DO data. It is likely that the detailed diffusion processes are different in each case. This is corroborated by the anomalously long response time of 63 s in humidified gas as shown in Fig. 7.7. Clearly further investigation is needed to clarify these issues.

7.5 Temperature dependence of quenching

Most sensors have to be corrected for temperature effects. In this work, both the absolute fluorescence intensity and the fluorescence quenching are temperature dependent. A study was carried out to investigate the effect of temperature on the performance of a DO sensor based on a R=2 MTEOS film.

This temperature dependence study was carried out using the 9 mL characterisation cell along with the gas calibration system shown in Fig. 5.9. Solutions of oxygenated and deoxygenated water were heated using the heater on the magnetic stirrer, and were then circulated through the measurement cell at temperatures ranging from 22°C to 40°C. The absolute fluorescence intensity in deoxygenated water was monitored, as was the quenching response, Q_{DO} . The fluorescence quantum efficiency decreases with increasing temperature.^{4, 5} Referring to Eqn. 2.1 in Chapter 2, the non-radiative decay rate increases with temperature at the expense of the radiative decay rate giving rise to the decrease in intensity as a function of temperature observed in Fig. 7.8 for R=2 MTEOS film. The temperature dependence of the quenching response will also include the dependence on temperature of the diffusion coefficient of oxygen in water and also in the sol-gel film. On its own, this would give rise to an increase in Q as a function of increasing temperature as D , the diffusion coefficient increases with temperature. Previous gas-phase oxygen studies have reported a decrease in absolute intensity and an increase in Q with increasing temperature as expected.⁴ The results of our study are shown in Fig. 7.8, where both absolute fluorescence intensity and Q_{DO} are plotted as a function of sample temperature.

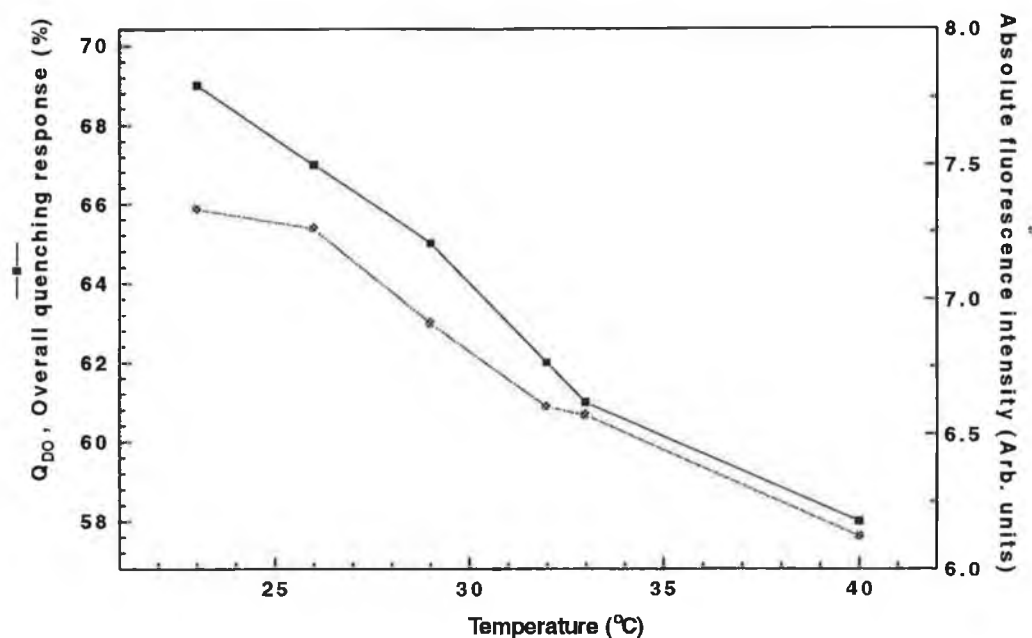


Figure 7.8: Overall quenching response in DO and the absolute fluorescence intensity in deoxygenated water as a function of water temperature.

It is clear from this diagram that, unlike the absolute intensity, the temperature dependence of Q_{DO} for our sensor does not behave as predicted. The quenching response, Q_{DO} , was found to decrease with temperature and to follow closely the decrease in absolute intensity. It is thought that this may be due to the temperature dependence of the oxygen solubility in water. Referring to Fig. 1.1⁶, extrapolation of this graph predicts an oxygen solubility of approximately 4 ppm at 40°C compared to 9.2 ppm at 20°C. This lower oxygen concentration would account for a lower Q_{DO} value at the higher temperature, and the solubility effect would appear to dominate the effect of temperature on the diffusion coefficient. To date, references to temperature dependence of only gas-phase quenching-based sensors have been found in the literature. The details of the temperature dependence of Q_{DO} are still under investigation in this laboratory. However, the observed temperature dependence is reproducible and can therefore be compensated for by the incorporation of an independent temperature sensor in the sensor head. This feature is being incorporated in the prototype design at present but its performance is beyond the scope of this thesis.

7.6 Limit of Detection and Sensor Resolution

Limit of detection (LOD) and sensor resolution are important quantities to investigate when evaluating the performance of a sensor. The limit of detection of the sensor in this case is based on three times the standard deviation at zero concentration ($\text{LOD} = 3\text{SD}$), and was calculated for the optimum sensing films, namely R=2 MTEOS and R=2 ETEOS films. The sensor resolution⁷ is the smallest increment in the value of the measurand that results in an unambiguous detectable increment in the output which is again based on three times the standard deviation. These data are tabulated in Table 7.3 below. The LOD is calculated from the slope of the Stern-Volmer plot shown in Fig. 7.3, at low oxygen concentrations (< 4 ppm), whereas the sensor resolution applies to the linear part of the graph from 5 to 40 ppm. This accounts for the differences between the values for LOD and sensor resolution, shown in Table 7.3, which would be the same if the slope of the plot were linear between 0 and 40 ppm.

	LOD	Sensor Resolution
R=2 MTEOS	15 ppb	0.23 ppm
R=2 ETEOS	5 ppb	0.18 ppm

Table 7.3: Values for sensor resolution and limit of detection for the optimum sensing films.

The values for LOD are excellent, and compare well with commercially available oxygen sensors based on polymer membranes. These are tabulated in Table 7.4 for comparison purposes.

Oxygen Sensor	LOD	Sensor Resolution
M-Squared Technology Ltd., Hampshire.	5 ppb	0.1 ppm / 0.01 ppm
Joanneum Research, Austria	-	0.1 ppm / 0.01 ppm

Table 7.4: Specifications from commercially available polymer-based optical oxygen sensors.

The LOD values for the sensing substrates, in particular that of the R=2 ETEOS films, are very promising for applications such as sludge monitoring, aeration control or corrosion monitoring, where it is necessary to monitor DO levels of parts per billion.

The sensor resolution appears to be unusually high, but as discussed previously, the sensitivity is different to that of the LOD due to the region of the plot used to calculate the sensor resolution. Very little averaging was used during the acquisition of this data, thus the signal-to-noise ratio could be improved by signal processing, which would increase the sensor resolution.

7.7 Low oxygen concentration (ppb) response

It was necessary to investigate the response of the sensor to low DO concentrations, as ppb resolution is required in applications such as sludge monitoring, aeration control, or corrosion monitoring. The system used to measure sensor response at low oxygen concentrations is shown in Appendix 1(iii). Calibrated DO solutions were achieved by using the standard mixing system of oxygen and nitrogen gases, mass flow controllers (MFC's), a water reservoir and a magnetic stirrer. The MFC's used in this case were a nitrogen MFC with a flow rate range of 0-500 sccm and an oxygen MFC with a flow rate range of 0-10 sccm. This ensured that extremely low DO solutions were achieved by appropriate mixing of the gases. These DO solutions were delivered to the cell for measurement via stainless steel tubing to prevent gas exchange with ambient air. For this reason the system was rearranged whereby the peristaltic pump was placed at the outlet of the cell such that the solutions were pulled through the system, and did not pass through polymer tubing until after the measurement had taken place. The sensing films used were R=2 MTEOS and R=2 ETEOS films.

In theory, this experiment appears practical and straightforward. In practice, however, achievement of stable low DO solutions was not trivial. It is obvious from data shown in Figs. 7.1 and 7.2, and LOD measurements in the previous section that these sensing films have very good resolution in low DO concentrations. The lowest DO concentration solution which was thought to have been obtained was a 80 ppb solution. It was not possible to verify this concentration as the resolution of the Clark electrode used by us is not good enough. Furthermore, due to the

instability of the solution, the Winkler method was a totally impractical measurement method. Fig. 7.9 shows some calibration data at low oxygen concentrations for a R=2 ETEOS films.

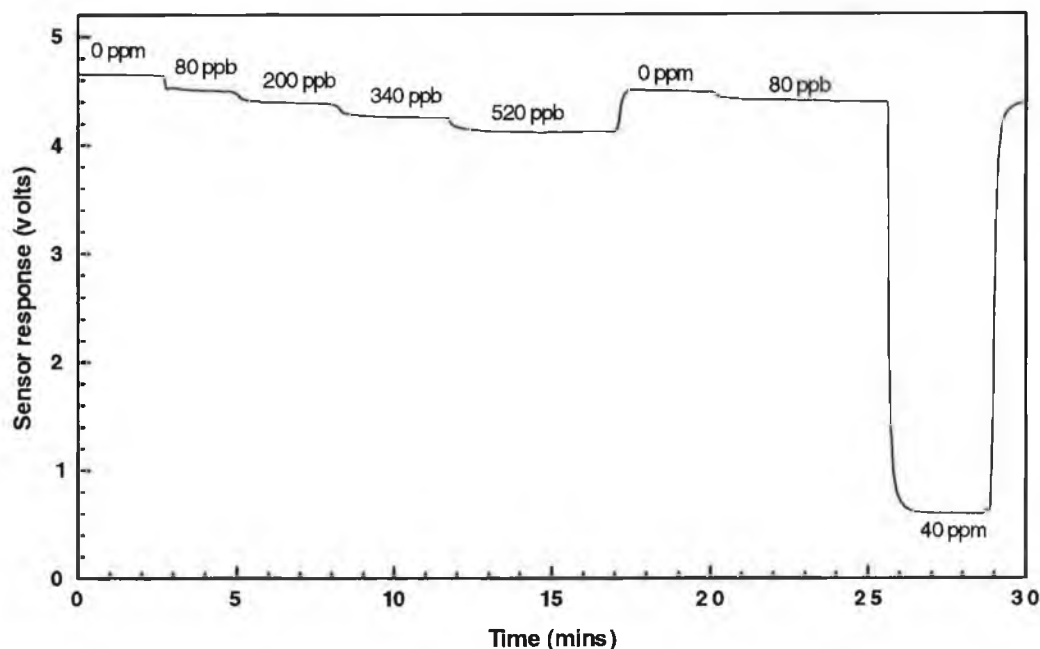


Figure 7.9: Response of R=2 ETEOS film to low DO concentrations.

Because of the small fraction of oxygen dissolved in these samples, the concentration was not stable. From the data obtained, it appears that oxygen in the air above the solution began to oxygenate the solution causing a gradual decrease in fluorescence intensity. As previously shown, this decrease is not due to an instability in the film. Nonetheless these data show that the LOD is much lower than 80 ppb, and that DO concentrations in the low ppb range are measurable.

7.8 Long-term stability and repeatability

Calibration data for the optimum sensing films, R=2 MTEOS and R=2 ETEOS, were presented in Section 7.2. Since the fabrication of these films, their quenching response in dissolved oxygen has been monitored and recorded periodically, over a period of 5 to 8 months. Figure 7.10 shows the overall quenching response, Q_{DO} , of the optimum films as a function of time after drying.

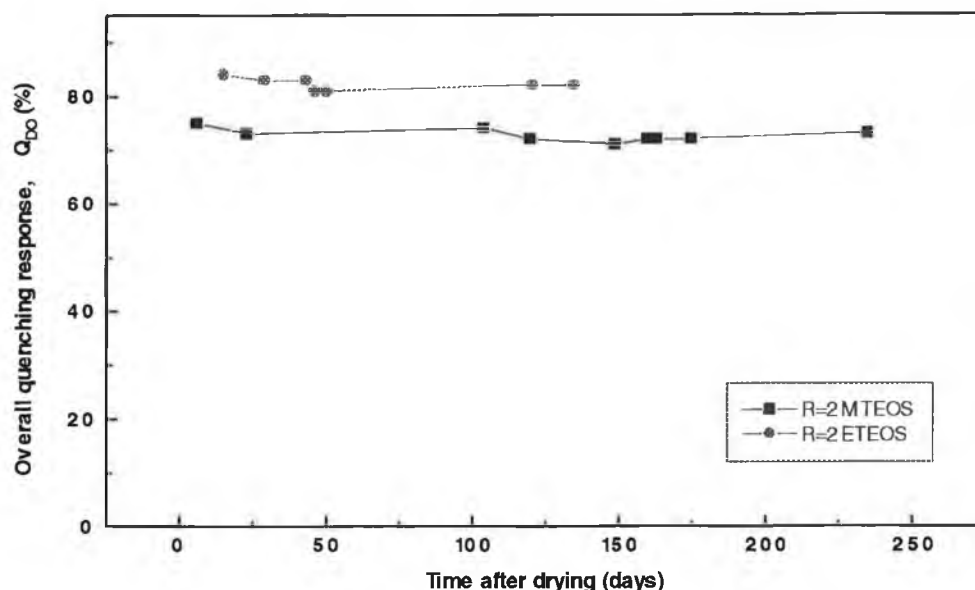


Figure 7.10: Overall quenching response, Q_{DO} , for R=2 MTEOS and R=2 ETEOS films as a function of time after fabrication

The slight variations are thought to be due to the characterisation system and lack of reproducibility in positioning the slide. As the overall quenching response in oxygen saturated water at room temperature has remained constant, the Stern-Volmer relationship (Eqn 2.12), would predict that the sensor calibration should also remain constant. Whether or not this includes the low oxygen concentration behaviour has not been verified.

Between these discrete measurements, the films were stored in ambient conditions in the laboratory, and were not stored in a dark, air-tight container. Since ultimately sensors based on these principles may be used for continuous DO monitoring, it is important that the slides are stored in water, either static or agitated, between discrete measurements, to measure their stability under more realistic conditions. Another stability test to be performed will involve continuous monitoring of slides in water, to ensure that their performance is not affected by continuous exposure to light.

7.9 Barrier layer

In anticipation of the deployment of sensors such as these in non-laboratory environments whose surface fouling and interferents could cause problems, the

use of a barrier layer coating on top of the sol-gel sensing film was investigated. An appropriate barrier would exclude any species which might interfere with the measurement process, and this layer might also minimise fouling. It is also important, however, that such a barrier layer have a high permeability to oxygen, for example, Nafion, silicone rubber and Teflon. In this work both Teflon and silicone rubber were tested. The Teflon used was Teflon AF-1601 (DuPont Fluoroproducts, DE., U.S.A.), and the procedure outlined in No. 2 below was specified by the manufacturer. The investigation was carried out as follows:

1. The response of R=2 MTEOS, R=2 ETEOS:TEOS 1:1, and R=2 TEOS films were measured in gas-phase oxygen and DO.
2. The films were then dip-coated with Teflon AF-1601, and the drying procedure was carried out as follows:
 - (i) Dry @ 50°C for 5-10 minutes
 - (ii) Dry @ 125°C for 5-10 minutes
 - (iii) Dry @ 165°C for 5-10 minutes
3. The response of the Teflon-coated films were then measured again in order to evaluate the impact on sensor performance, before examining the effect of interferents.

Table 7.5 shows the values of Q_{DO} for the three films tested.

	Q_{DO} Before coating	Q_{DO} After coating
R=2 TEOS	20%	20%
R=2 ETEOS:TEOS 1:1	72%	81%
R=2 MTEOS	73%	86%

Table 7.5: Values for Q_{DO} of Teflon-coated films.

Both ormosil films exhibit enhanced quenching after coating. R=2 MTEOS films showed the greater enhancement, with Q_{DO} increasing from 73% to 86%. Q_{DO} for R=2 ETEOS:TEOS 1:1 films was also enhanced, but Q_{DO} for R=2 TEOS films did not change at all. It was found, on removal of the TEOS slide from the characterisation cell, that the Teflon coating had cracked and peeled off the slide.

This prompted an investigation of the quality of the Teflon coating on the other slides, which revealed that the coating on R=2 MTEOS films seemed to adhere well, but the coating on R=2 ETEOS:TEOS 1:1 film rubbed off when moist.

There is clearly an adhesion problem with Teflon particularly on TEOS films. Also the signal-to-noise ratio (SNR) is much lower after coating the films with Teflon. This may suggest that the films are not homogeneous, and that light is caused to scatter, resulting in a lower SNR.

Silicone rubber was then investigated. The silicone rubber used was a medical grade Dow Corning 3140 RTV. A 50% w/v solution of silicone rubber in xylene was used to coat TEOS-based and organically modified sensing films. They were coated using a slow dip-speed to achieve thin barrier films, and were left to cure in a fumehood for a number of days. The DO sensing performance of these films after coating is summarised in Table 7.6.

As can be seen, TEOS-based films showed no enhancement when coated with silicone rubber. This is probably due to humid gas permeating the silicone barrier layer, and condensing on the hydrophilic TEOS film, thereby reducing the total quenching effect as discussed previously in Section 6.2.2.1. The ormosil sensing films do, however, exhibit minor enhancement after coating with silicone rubber. Although as seen in Table 7.5, Teflon-coated films exhibit a larger quenching response than silicone-coated films, silicone rubber was chosen as the barrier layer for interferent studies discussed in Section 7.10.

	Q_{DO} Before coating	Q_{DO} After coating
R=2 ETEOS	80%	81%
R=2 MTEOS	73%	75%
R=2 MTEOS:TEOS 2:1	62%	65%
R=2 TEOS	20%	20%
R=4 TEOS	6%	6%

Table 7.6: Q_{DO} for modified films before and after coating with silicone rubber.

Generally the film quality for silicone rubber films was superior and the SNR was also greater. Silicone rubber is widely available and has a higher permeability to

oxygen than Teflon. Films used in the study of interferences were R=2 MTEOS and R=2 ETEOS films, uncoated, and coated with a thin layer of 50% w/v solution of silicone rubber (3140 RTV, Dow Corning) in xylene.

7.10 Interference tests

Interference tests were carried out using the system shown in Fig. 5.11. An initial test was performed on uncoated R=2 MTEOS and R=2 ETEOS films. The quenching response of these films was measured using tap water rather than deionised water, and Q_{DO} was found to be unchanged for both films. This was quite a promising result, as tap water contains many ions such as magnesium (Mg^{2+}), calcium (Ca^{2+}), carbonates (CO_3^{2-}), chlorides (Cl^-) and fluorides (F^-), and these did not interfere with oxygen quenching. Tap water was then used as the reference solution. The interferent solution being tested was contained in the second reservoir. Both reservoirs were exposed to ambient conditions such that the solutions were aerated and therefore contained approximately 9 ppm of oxygen at standard temperature and pressure. The main interferents tested were heavy metal ions and surfactants. Heavy metal ion solutions of concentration 20 ppm were made using metal salts of magnesium, copper, zinc and aluminium. The surfactants used were 0.1 mM solutions of cetyl trimethylammonium bromide (CTAB) and sodium dodecyl sulphate. These solutions, along with the reference solution, were left to equilibrate at ambient temperature and pressure, so that the DO concentration was equal for both solutions, after which the fluorescence intensity of the sensor in the reference solution was compared with that in the interferent solution. The results of these measurements were found not to be repeatable. If the interferent tests were carried out after the solutions were left for several hours or overnight in ambient conditions, little or no interference took place. However, if these tests were repeated having left the solutions to equilibrate for only a short period of time, some interference would take place. It is thought that this was due to differences in oxygen concentrations between the two solutions. As a result, the experimental system was modified such that the solutions were deoxygenated using nitrogen degassing to ensure that no oxygen was present to quench the ruthenium complex. Klimant and Wolfbeis⁸ in their study of interferences on a silicone-based oxygen sensor carried out the study

using nitrogen-purged solutions. Using this modified system little or no interference occurred as can be seen in Table 7.7.

Possible Interferents	Concentration	Signal change R=2 MTEOS	Signal change R=2 ETEOS
Copper	20 ppm	< 2%	< 1.5%
Aluminium	20 ppm	< 1.5%	< 1%
Zinc	20 ppm	< 1%	< 1%
Magnesium	20 ppm	< 1%	< 1%
CTAB	0.1 mM	< 2%	< 1.5%
Sodium dodecyl sulphate	0.1 mM	< 2%	< 1.5%

Table 7.7: Effect of potential interferents on the fluorescence intensity of uncoated MTEOS and ETEOS films in nitrogen-purged water.

Both uncoated and silicone-coated MTEOS and ETEOS films were tested. None showed significant interference effects. If the bare sensing films had shown some interference by, for example, ionic quenchers, coating with a hydrophobic barrier layer such as silicone rubber, would exclude these substances and prevent them interfering with the quenching process.⁸ It is possible that a thin barrier layer may act as a protective layer without compromising the response time.

7.11 Conclusions

The performance of films prepared using the optimal sensor preparations identified in Chapter 6 was reported in detail in this chapter. In particular, sensor calibration curves, temporal stability, sensor resolution, LOD and response time were examined. These were found to compare well with those of commercially available sensors. Temperature effects and response to interferents were also investigated. The overall performance of the two optimal sensor films is summarised in Table 7.8.

	R=2 MTEOS	R=2 ETEOS
Q_{DO}	73 ± 2 %	80 ± 2 %
Q_{DO} stability	± 2% over 8 months	± 2% over 5 months
LOD	15 ppb	5 ppb
Sensor resolution	0.23 ppm	0.18 ppm
Response time	9 ± 3 s	17 ± 3 s

Table 7.8: Quantitative data indicating the overall performance of the optimum sensor films.

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CHAPTER 8:

RESULTS FROM DIFFERENT SENSOR CONFIGURATIONS

8.1 Introduction

In this chapter, evolution of the experimental characterisation system for the planar DO system is described. The developments are traced from an initial laser-based system to the current all-solid-state unit incorporating a small volume (1 cm^3) glass cell. Chapter 6 concentrated on the optimisation of sensing films for DO sensing, whereas this chapter emphasises the improvement in results as the instrumentation is updated and optimised. Alternative sensor configurations such as an optical fibre-based system and a phase fluorimetric system are also described. Fibre-based sensor data are reported and the suitability of this evanescent wave configuration is demonstrated. Phase fluorimetry has many advantages associated with it for this application. Early gas phase results, as well as modulation frequency optimisation data and phase fluorimetric DO sensing results are presented.

8.2 Evolution of characterisation system for planar DO sensor

Planar sensing substrates were prepared for coating as described in Section 4.3. The coating sol fabrication process is detailed in Section 4.2. Initially a laser-based characterisation system was used to investigate the potential of the films for DO sensing. Having demonstrated this, it was necessary to design a more compact, portable system from which more repeatable results could be obtained.

8.2.1 Laser-based system

The laser-based characterisation system shown in Figure 5.2 was used for preliminary DO investigations. The sample was excited using the 488 nm line of the argon-ion laser, and the fluorescence was detected using a R928 photomultiplier tube, while the oxygen gas from the mass flow controller was flowing through the sample cell. The films fabricated with TEOS and having an R value of 2, showed excellent sensitivity to gaseous oxygen. During this investigation, as detailed in Chapter 6, it was found that by varying the R value, the calibration data varied, and consequently the films could be optimised to sense

gaseous oxygen in a particular concentration range.

Having seen how the sensitivity to oxygen gas could be tailored by varying the process parameters in the sol-gel fabrication process, the emphasis then shifted to sensing oxygen dissolved in water. As was seen in Chapter 1, the concentration of dissolved oxygen in water at 20°C and standard pressure is 9.2 ppm. This is a much smaller relative oxygen concentration compared with gas phase oxygen concentrations. The same laser-based system as was used for gas phase sensing was employed. A basic sample cell was assembled, whereby the sample was held rigidly while the gas flowed into the water cell. Initial DO experiments involved recording the fluorescence emission spectrum of $[\text{Ru}^{(\text{II})}(\text{Ph}_2\text{phen})_3]^{2+}$ -doped TEOS-based sol-gel films in deoxygenated water, aerated water and oxygenated water. The resulting data is shown in Fig. 8.1. Calibration data can be seen in Fig. 8.2.

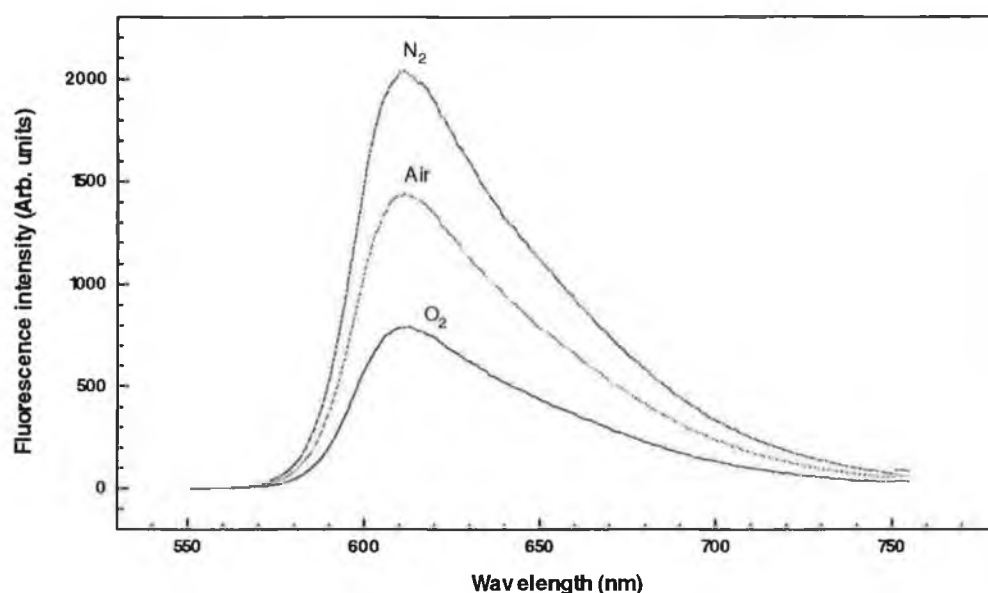


Figure 8.1: Ruthenium-doped sol-gel film fluorescence emission spectra for deoxygenated, aerated and oxygenated water.

These early results illustrate the potential of the doped-sol-gel-derived films for DO sensing. However, it is clear that materials development work, in conjunction with improvements in the instrumentation, need to be carried out to optimise the films for this application. The response time and noise values reflect the unoptimised nature of the characterisation system rather than the inherent behaviour of the sensing films.¹ The low signal-to-noise ratio is due in part to light scatter from gas

being bubbled into the sample cell. This led to the development of a smaller, more stable characterisation system.

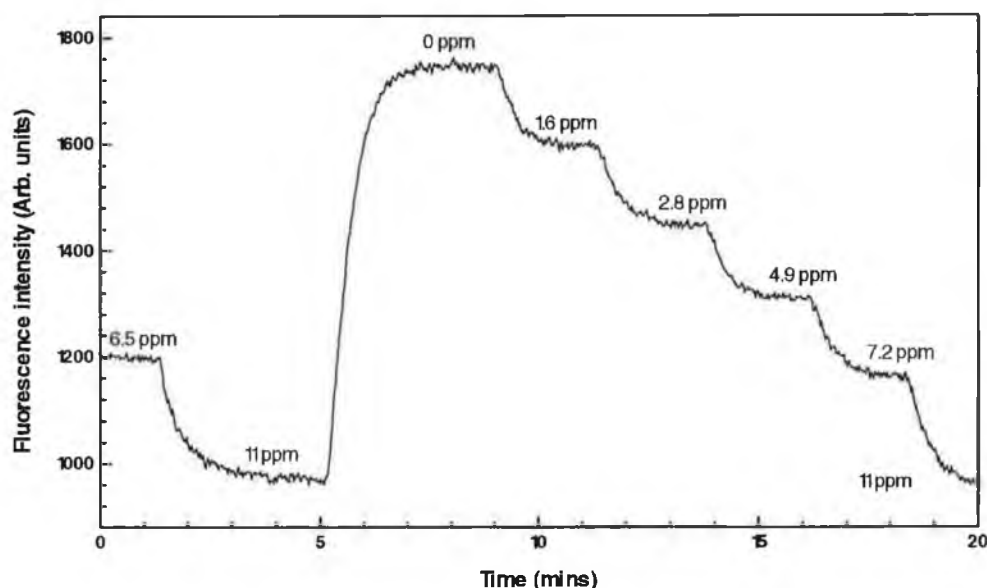


Figure 8.2: Real-time response of sol-gel film to different concentrations of DO.

8.2.2 All solid-state system

This system consists of three main parts; the oxygen/dissolved oxygen cell, the circuitry and the optics. A schematic diagram of the compact all-solid state characterisation system can be seen in Fig. 5.6, the circuit diagrams for the electronics are reproduced in Appendix 2, and the optics, for example, filters and lenses are detailed in Section 5.3.1. The circuitry incorporates a balanced modulator/demodulator which, through synchronous detection with the LED pulsing signal, permits effective lock-in detection and discrimination against ambient light. The filters were chosen such that the detection of excitation light is minimised, together with minimal reduction of the fluorescence signal. The sensing substrate is held firmly in the lid of the cell and the optics are mounted in a stable configuration on the main frame. It is a totally self-contained system which yielded reproducible data, allowing direct comparisons between different sensing films. An increased signal-to-noise ratio (SNR) was achieved because the calibration solutions are mixed in a reservoir, and not in the cell, after which they are pumped into the cell using a peristaltic pump. This minimises the scatter due

to bubbles in the system.

Fig. 8.3 shows the quenching response of a R=2 TEOS film in dissolved oxygen using the first all-solid state calibration system (Sample cell volume = 23 cm^3), which is shown in Appendix 1(i). The response has greatly improved in comparison with the response of the films obtained using the laser-based system (Fig. 8.2). It was at this stage in the development of the characterisation instrumentation that the investigation of organically modified sensing films, as described in Chapter 6, began.

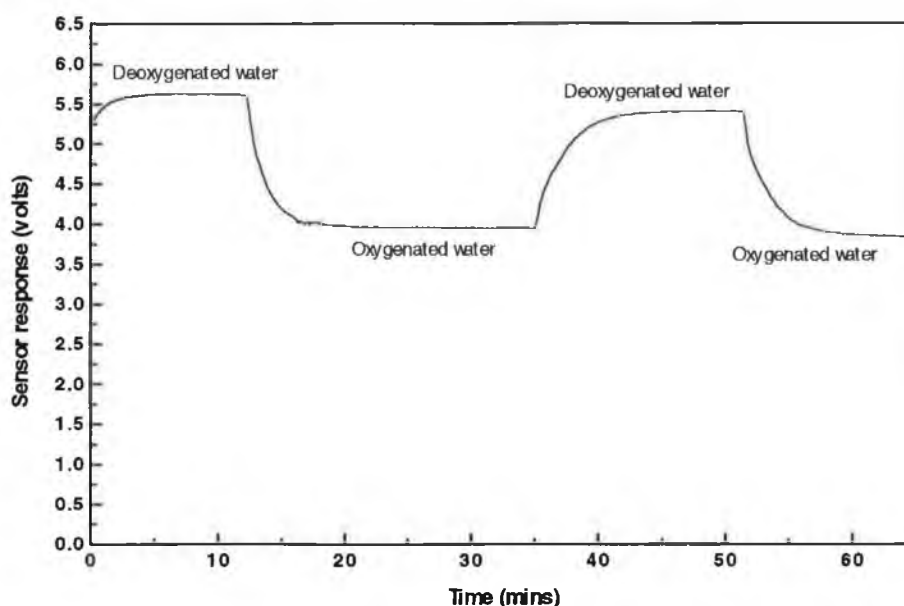


Figure 8.3: Quenching response of R=2 TEOS film in DO using larger characterisation cell.

A further improvement resulted from the fabrication of a smaller volume characterisation cell, which can be seen in Appendix 1(ii). The main difference between this and the previous characterisation cell is size. The volume of this smaller cell is 9 cm^3 , which is less than half the size of the first cell. The quenching response of a TEOS film using this cell can be seen in Fig. 8.4. TEOS-based films are shown here so that a consistent comparison of the cells can be undertaken. An obvious difference between Figs. 8.3 and 8.4 is the shorter system response time of the latter. This reflects the change in delivery time, fill time and gas diffusion. In Section 7.3, the topic of response time was discussed, and it was found necessary to reduce the cell volume to the order of the size of the

sensing substrate in order to get an approximate value of the response times of the films.

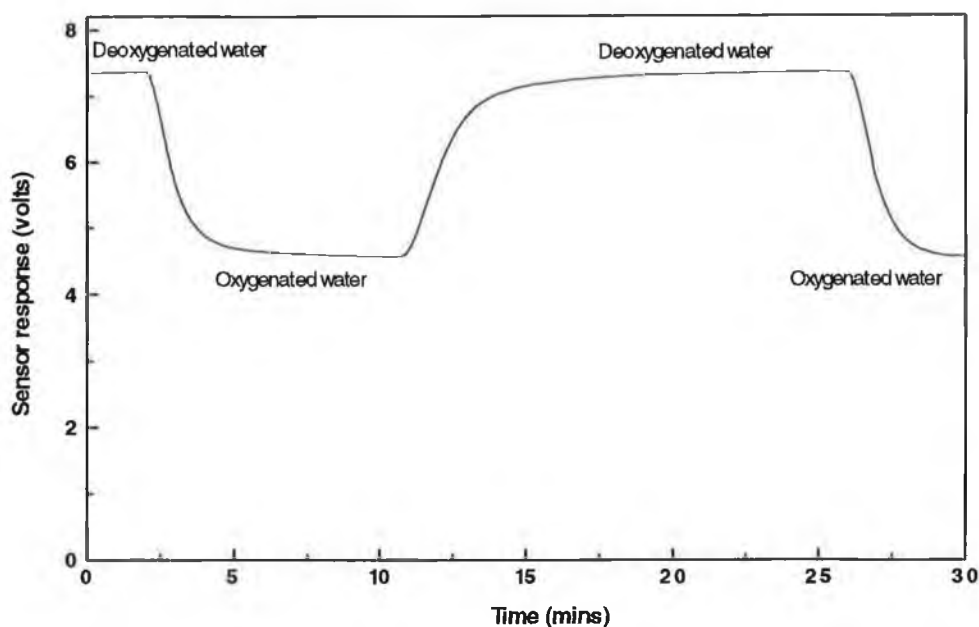


Figure 8.4: Quenching response of R=2 TEOS film in DO using smaller characterisation cell.

The third and smallest sample cell is made of glass and has an internal volume of approximately 1 cm^3 . Diagrams of this cell can be seen in Appendix 1(iii). As well as having the advantage of being able to estimate the response times of the films, the reduced volume enables faster fill times which speeds up the scan time. Graphs of the fast response times using the microlitre cell for optimum films are shown in Fig. 7.4 and 7.5.

The characterisation system for scanning planar substrates has been optimised such that the response of planar films to oxygen dissolved in water can be determined quickly, easily and repeatably, thus enabling the acquisition of calibration data for various sensing films.

8.3 Fibre-based intensity DO sensor

The planar-based fluorescence intensity system was extended to develop a fibre-based system, which could be developed as a dipstick-type probe. The design used here was based on a gas-phase fibre sensor developed previously in this

laboratory.^{2, 3} The sensing mechanism is via evanescent-wave interactions with coatings on a de-clad optical fibre. Plastic clad silica (PCS) fibres with a core diameter of 600 μm , were dip-coated with a ruthenium-doped organically modified sol-gel film as described in Chapter 4. A diagram of this experimental set-up is shown in Fig. 5.7. The electronics used in this set-up were exactly the same as those used in the planar system and are shown in Appendix 2, but the optics are arranged differently such that the fluorescence is collected at the proximal end of the fibre. This improves the signal-to-noise ratio, as 90% of the excitation light exits out the distal end of the fibre.³ When the excitation light passes through the blue filter, it is incident on a dichroic filter, which is at an angle of 45° to the incident beam. This filter reflects the 450 nm excitation light which is then focused into the fibre and the evanescent wave excites the ruthenium complex in the sol-gel film. The fluorescence from the fibre is transmitted by the dichroic filter, and is focused onto a photodiode detector.

The sensing films used in this work are R=4 MTEOS films, because of their fast stabilisation times. Fig. 8.5 shows the overall quenching response of these films in DO.

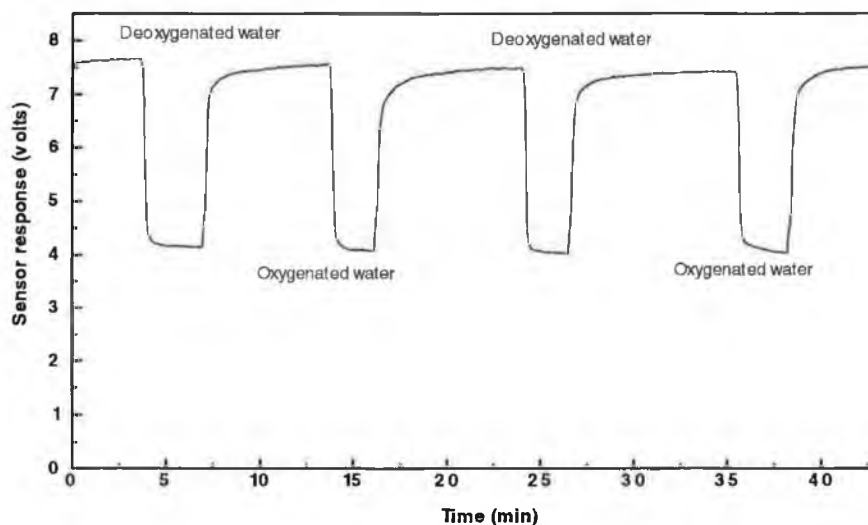


Figure 8.5: Overall quenching response of R=4 MTEOS fibre-coated films in DO.

Calibration data for the fibre optic system are shown in Fig 8.6, and the corresponding Stern-Volmer plot can be seen in Fig. 8.7.

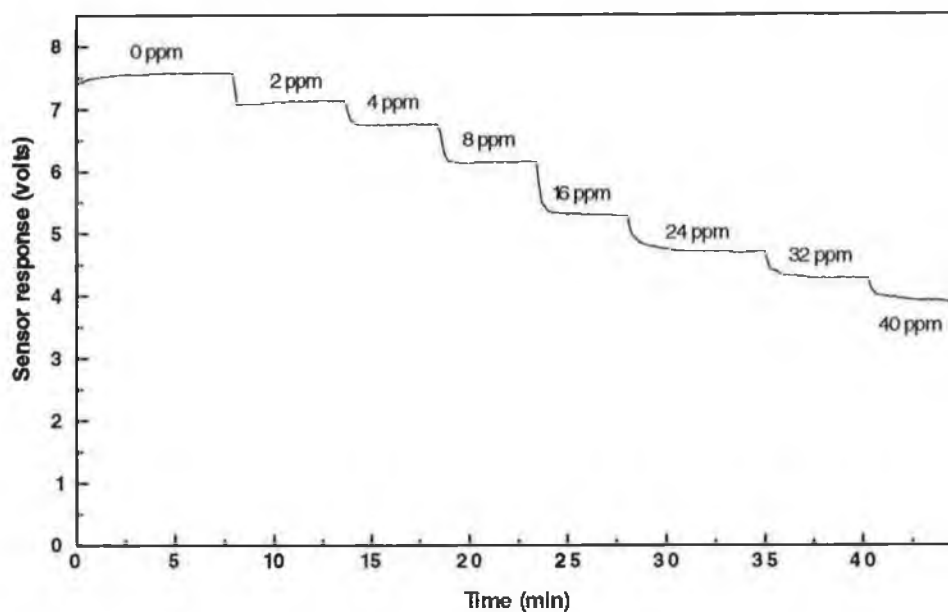


Figure 8.6: Calibration data for R=4 MTEOS fibre-coated sol-gel film in DO.

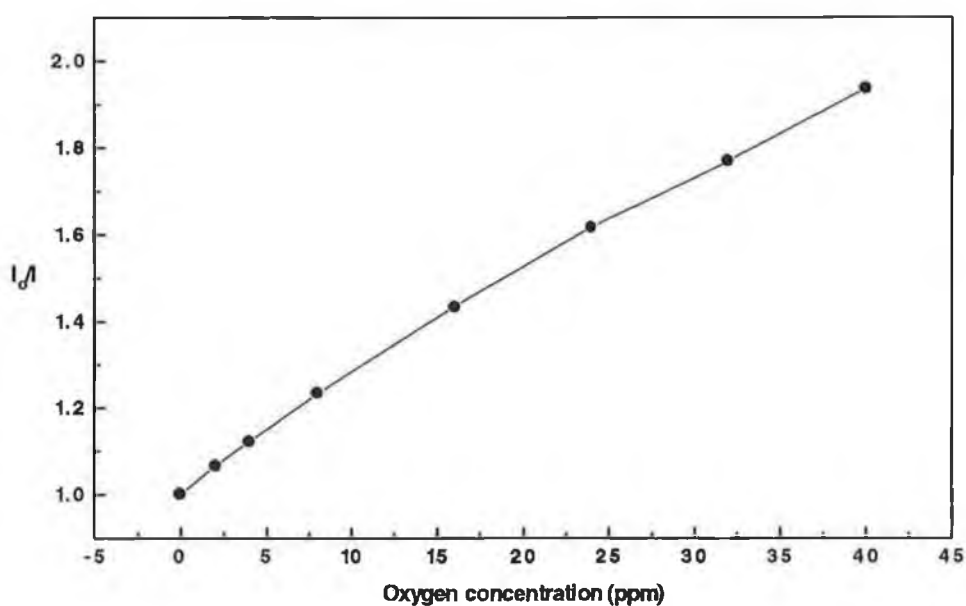


Figure 8.7: Stern-Volmer plot of R=4 MTEOS fibre-coated film in DO.

The overall quenching response of R=4 fibre-coated MTEOS films in DO is 46%., This is a much lower value than the value for corresponding planar-coated films. Q_{DO} for R=4 MTEOS planar-based films is 64%. The origin of this discrepancy is

not clear. Due to the cylindrical-shape of the optical fibre, it is possible that the drying regime is very different when compared with that of the planar films. The fibre geometry may result in the deposition of thinner films, and these films may dry more quickly than planar films, causing reduced porosity, and hence a reduced overall quenching response. Other researchers have also found variations of Q_{DO} and K_{SV} with fabrication conditions.⁴

The results shown in this section demonstrate the potential of a fibre-optic DO sensor. This fibre-optic sensor can be used in a probe configuration, but as it stands is not rugged enough to withstand harsh conditions, such as turbulent water in aeration control, where there is a risk that the fibre sensor may break. Nonetheless, it is possible to use this sensor as an *in situ* sensor for example in corrosion monitoring. It may also be used in remote or distributed sensing⁵, where a number of analytes could be detected, for example DO, pH or ammonia.

8.4 Lifetime analysis

Lifetime-based sensors have inherent advantages over intensity-based sensors as discussed in Section 2.5, such as they are not affected by static quenching, measurements are independent of fluorophore concentration, and no drift results from light source intensity and photodetector sensitivity fluctuations as it is not absolute intensity that is measured but rather time lag between excitation and fluorescence.^{6,7} As part of preliminary work on this development, lifetime analysis was performed on doped sol-gel films in gas phase to investigate the variation of lifetime, τ , with oxygen concentration, as shown by the Stern-Volmer relationship for lifetimes,

$$\frac{\tau_0}{\tau} = 1 + K_{sv} [Q] \quad (2.13)$$

The system used for lifetime analysis is shown in Fig. 5.4. The sol-gel coated fibre substrates were excited using 15 ns pulses from a frequency-doubled Nd:YAG laser ($\lambda = 532$ nm). The coated substrates were mounted in a gas cell, and a range of oxygen gas concentrations was achieved by precise mixing of 100% oxygen and 100% nitrogen, respectively. The decays were saved using a digitising storage oscilloscope, and downloaded to the computer via a data acquisition package, Scopelink (Hewlett Packard). Fig. 8.8 shows typical

fluorescence decay curves for the sol-gel-immobilised ruthenium complex in the presence of 100% nitrogen and 100% oxygen, respectively.^{7,8}

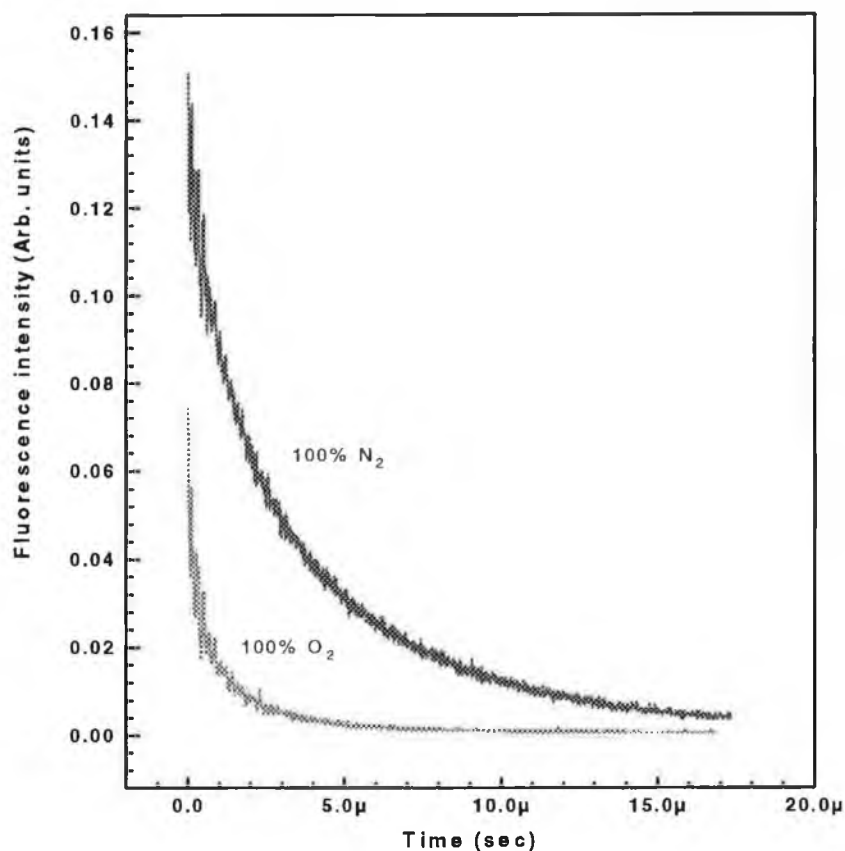


Figure 8.8: Sol-gel immobilised ruthenium fluorescence decay curves.

The observed fluorescence decays are not single exponential. The lifetime data measured at a range of concentrations from 0% to 100% oxygen were analysed, and in all cases the data provide an approximate mathematical fit to a double exponential decay. This behaviour is consistent with a two-site model for the ruthenium complex, where the two sites have different levels of accessibility to oxygen. Alternatively, this behaviour may represent an approximate fit to a continuous distribution of lifetimes due to a heterogeneous sol-gel environment. For each oxygen concentration, long and short lifetimes were determined. In 100% nitrogen, values of 5.613 μs and 933 ns, respectively, were found to give the best fit. Long and short lifetime components of 2.217 μs and 356 ns were found for 100% oxygen. Pre-exponential factors were also determined from the analysis. This investigation was carried out using R=2 TEOS films.

Lifetime-based sensing has been shown to be a viable method of characterising quenching behaviour in gas phase. These preliminary results from the laser-based system established the feasibility of a lifetime-based sensor for both gas phase and DO measurements. This is detailed in the next section.

8.5 Phase fluorimetry

In some applications, it may be advantageous to use a lifetime-based method of sensing. As mentioned in Chapter 2 and in the previous section, fluorescence lifetimes are immune to problems which may affect intensity-based measurements such as photobleaching and detector drift.^{6, 7} Although, no problems of this nature affected the DO intensity-based sensor, this method of sensing was investigated to examine the potential of applying it to DO sensing should the need arise. The specific technique used by us is phase fluorimetry. When a fluorescent material is excited by sinusoidally modulated light, the resulting fluorescence is phase shifted relative to the excitation signal by an amount that is dependent on the lifetime of the fluorescent complex. In this manner, the concentration of the quencher, in this case oxygen, can be deduced by measuring the phase shift of the emission relative to the excitation signal.

Theoretical analysis of lifetime data was carried out initially in order to identify an optimum modulation frequency. The results of this analysis were then compared with experimental measurements. Following this, a dedicated phase fluorimetric DO sensor system was constructed and characterised.

8.5.1 Phase fluorimetric oxygen gas sensing

This work was performed in conjunction with the development of a phase fluorimetric oxygen gas sensor in this laboratory.^{8, 9} The measured lifetime data obtained in Section 8.4 was used to calculate the expected phase shift for any oxygen concentration for a given modulation frequency using Eqns. 2.22 and 2.24. The phase shift will be smallest for a concentration of 100% oxygen and greatest for a concentration of 0% oxygen. Phase shifts were calculated for 100% nitrogen and 100% oxygen gas at various modulation frequencies. The calculated phase shift difference between these two extremes of the measurement range, ϕ_{diff} ,

where $\phi_{\text{diff}} = \phi_{\text{nit}} - \phi_{\text{oxy}}$, is plotted as a function of modulation frequency in Fig. 8.9. From the diagram, it is noted that a maximum phase difference of 27° is predicted to occur at 48 kHz, and that the response levels off close to 60 kHz.

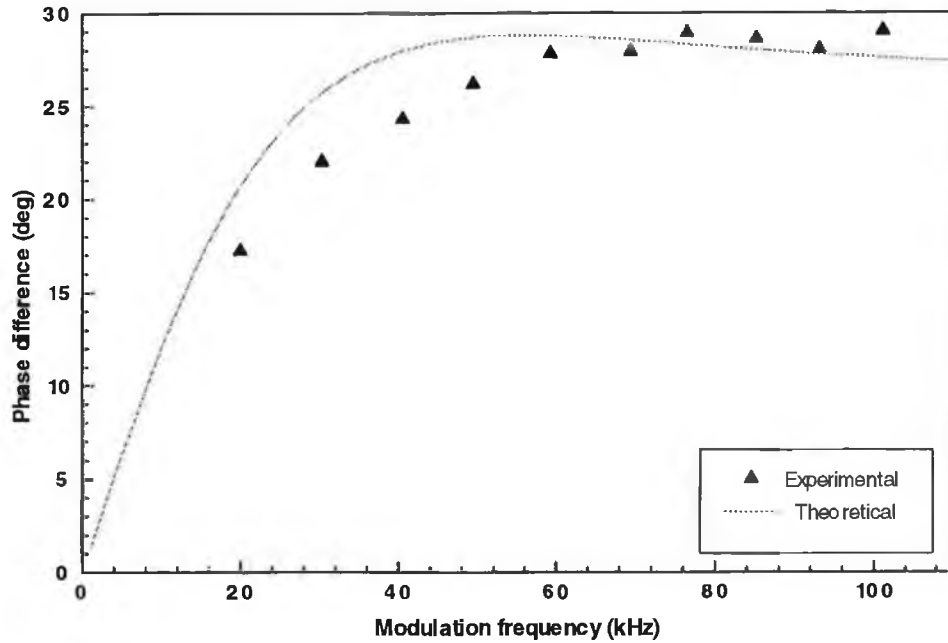


Figure 8.9: Theoretical and experimental phase difference, $\phi_{\text{diff}} = \phi_{\text{nit}} - \phi_{\text{oxy}}$ plotted against modulation frequency for gas phase.

The optimum modulation frequency for maximum phase shift was also determined experimentally to an accuracy of 0.1 of a degree, using the dual-phase lock-in amplifier and LED-based phase fluorimetric system for fibre-based gas phase measurements.³ The LED modulation frequency was varied by adjusting the LED driving frequency. For each frequency, the resulting phase shift was recorded as the sensor environment was alternated between 0% and 100% oxygen. These experimental phase difference results are also plotted in Fig. 8.9. Given the approximation inherent in the double exponential decay model used, these results are in reasonable agreement. The agreement is sufficiently close to enable selection of the optimum operating frequency range for the fibre-based phase fluorimetric oxygen gas sensing system.³

8.5.2 Dissolved oxygen phase fluorimetry

Section 5.3.3 described the instrumentation used for dissolved oxygen phase fluorimetry experiments.

An important factor in phase fluorimetry, as was seen in the previous section, is the choice of modulation frequency. Before any measurements are made, the optimum modulation frequency must be found. Fig. 8.10 shows a theoretical plot of demodulation factor, m , and phase angle, ϕ , as a function of $\log(\text{modulation frequency})$. The demodulation factor, m , is proportional to the sinewave modulated intensity as seen in Fig. 2.7, and subsequently the signal-to-noise ratio (SNR) decreases with m .

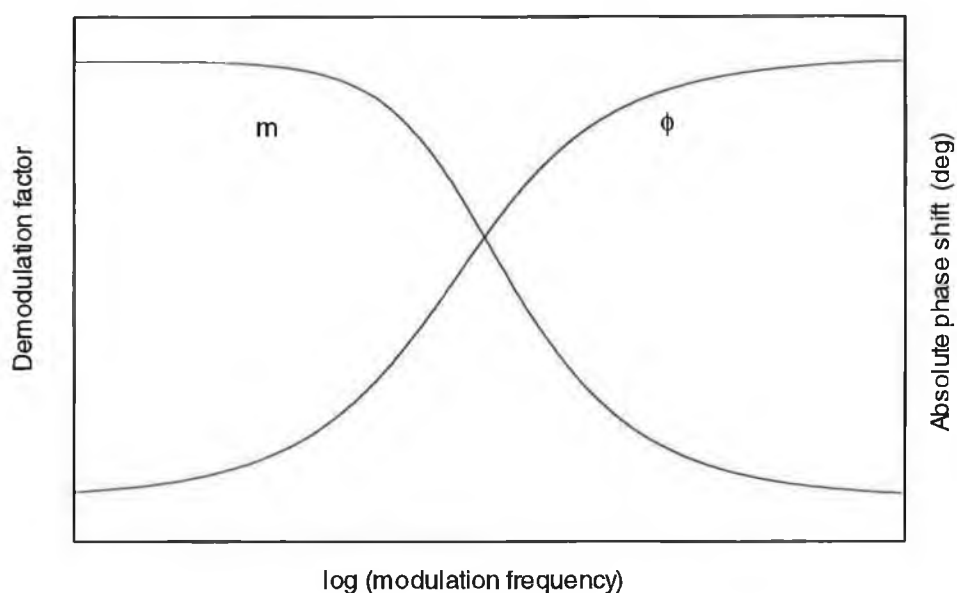


Figure 8.10: Phase angle and demodulation frequency as a function of $\log(\text{modulation frequency})$

As the $\log(\text{modulation frequency})$ increases, the demodulation factor, and hence the modulated intensity decreases, but the phase angle increases. Initial experiments were carried out to find the optimum modulation frequency. This was done by monitoring the phase angle in deoxygenated water and in oxygenated water while increasing the modulation frequency. As expected, the phase angle increased with increasing modulation frequency, but the modulated intensity as well as the signal-to-noise ratio decreased.

Fig. 8.11 shows the phase difference, ϕ_{diff} , where $\phi_{\text{diff}} = \phi_{\text{deoxy}} - \phi_{\text{oxy}}$, plotted against modulation frequency for R=2 MTEOS film in DO.

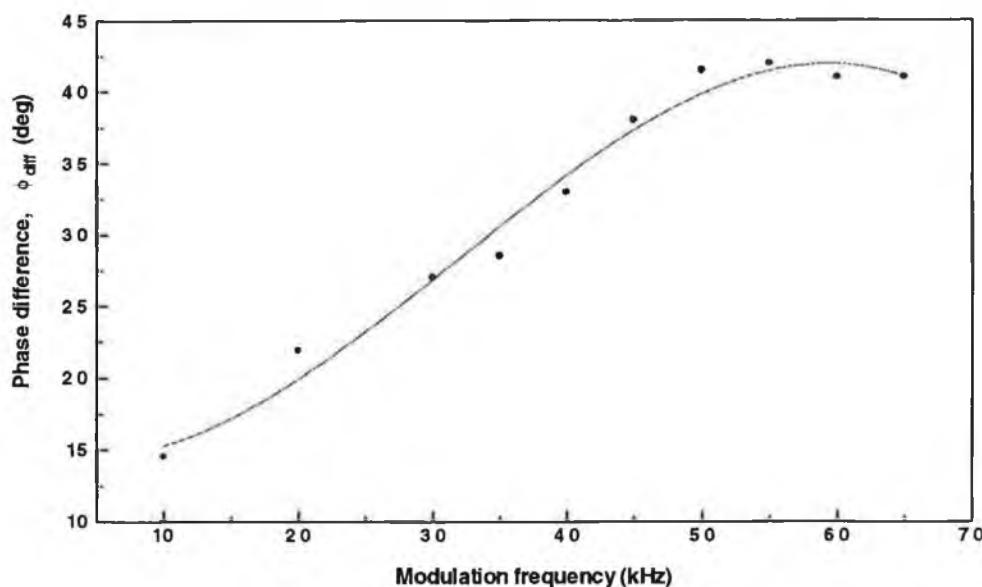


Figure 8.11: Phase difference, ϕ_{diff} , as a function of modulation frequency for R=2 MTEOS film in DO.

This behaviour is similar to Fig. 8.9 for gas phase. The phase difference increases steadily up to approximately 45 kHz, after which it levels off up to 65 kHz. After 45 kHz it became increasingly difficult to measure the phase difference because of the poor fluorescence intensity signal. As a consequence the optimum modulation frequency was found to be 45 kHz. Although it should be possible to increase the signal-to-noise ratio, by improving the detection electronics, this trade-off between fluorescence intensity and phase shift as seen in Fig. 8.10, will always be present. Nonetheless, the phase difference corresponding to the chosen optimum modulation frequency is approximately 45° compared with 27° for TEOS films in gas phase (Section 8.5.1). This reflects a change in fluorescence decay time in the MTEOS film. Using the optimum modulation frequency of 45 kHz, experiments were carried out to investigate the DO quenching response for the sensor film. The overall quenching response in DO of R=2 MTEOS films is shown in Fig. 8.12. These data which show a phase difference of just less than 45° , have been offset to yield a phase angle of 0° in fully oxygenated water (40 ppm).

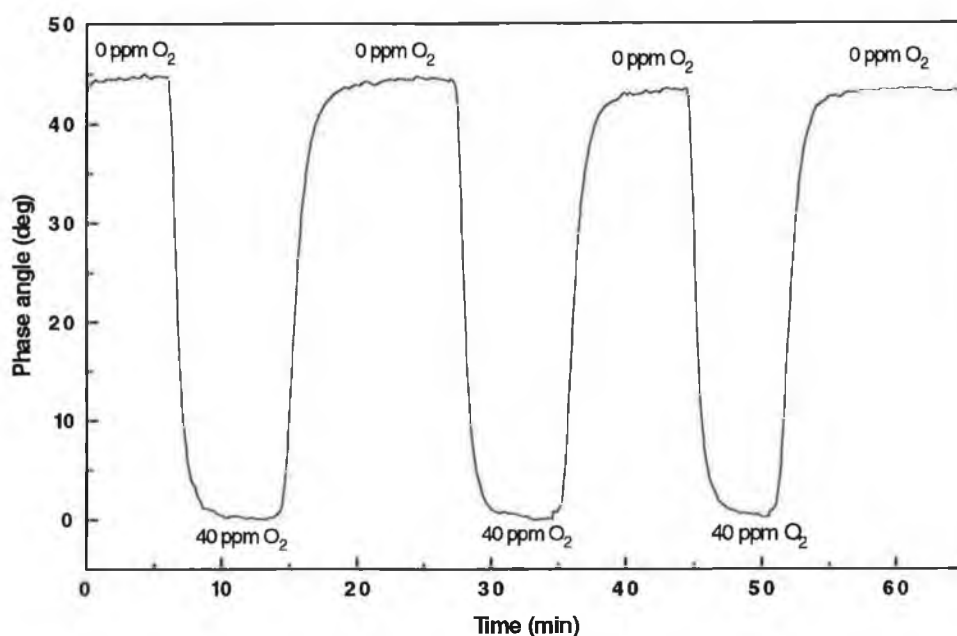


Figure 8.12: Real-time phase fluorimetric DO sensing recorded at a modulation frequency of 42 kHz.

Fig. 8.12 illustrates a number of important features of the sensor. It exhibits a high level of repeatability, and also exhibits high sensitivity between the extremes of the measurement range, which indicates that changes in the oxygen concentration can be measured with high resolution. The response time is limited by the size of the cell as discussed previously.

Calibration data for a R=2 MTEOS film in DO are shown in Fig. 8.13. As before, the sensor exhibits greatest sensitivity at lowest DO concentrations. The signal-to-noise ratio is quite a bit lower than that of fluorescence intensity measurements, as the system electronics have not yet been optimised. Dedicated electronics are being developed at present in the laboratory to optimise phase fluorimetric detection for both gas phase and DO sensing.

These preliminary phase fluorimetry data indicate the potential of using this technique for sensing dissolved oxygen. It is a viable detection method for sensing as the instrumentation can easily be miniaturised allowing portability of the sensor, as well as being a relatively inexpensive lifetime-based sensor.

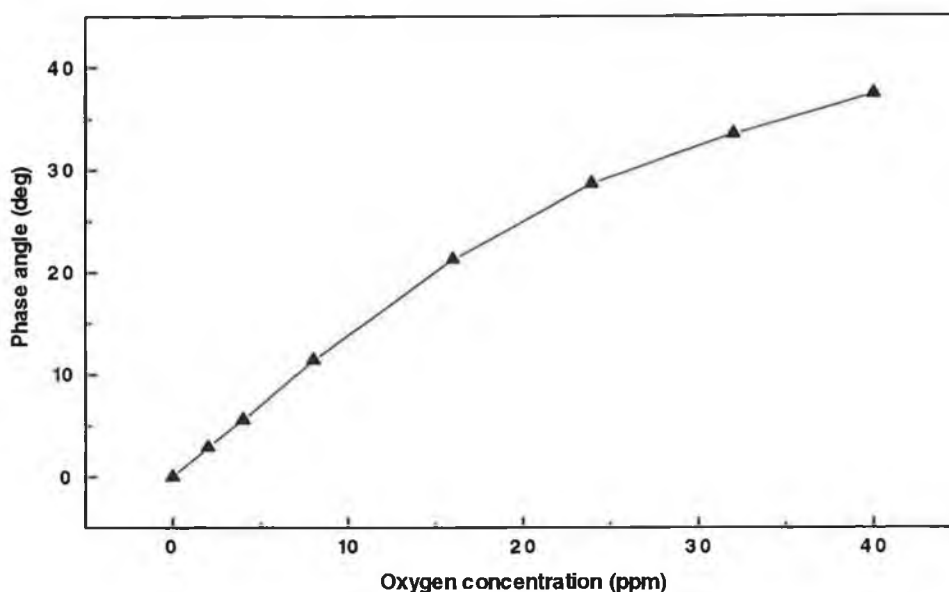


Figure 8.13: Calibration data for R=2 MTEOS film in DO using phase fluorimetry.

8.6 Conclusion

A number of sensing techniques, in conjunction with different sensor configurations have been demonstrated. The main work on the DO sensor has been performed successfully on planar substrates in fluorescence intensity mode. By reducing the volume of the sample cell, the scan time and fill time were reduced making it possible to evaluate response times of the sensors.

Other sensor configurations were also investigated. This led to the development of a fibre-based sensor in fluorescence intensity mode. These data show the potential of fibre sensors for monitoring dissolved oxygen.

Fluorescence lifetime sensing was shown to be a viable alternative to intensity sensing for DO, with many inherent advantages. This led to the demonstration of a DO phase fluorimetric sensor.

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9. G. O’Keeffe, B. D. MacCraith, A. K. McEvoy, C. M. McDonagh, J. F. McGilp; **Development of a LED-based phase fluorimetric oxygen sensor using evanescent wave excitation of a sol-gel immobilised dye**, *Sens. Actuators*, 1995, Vol. B 29, pp. 226-230.

CHAPTER 9: CONCLUSIONS

9.1 Objectives revisited

Overall the main objectives set out in Section 1.6 have been achieved. A commercially viable optical dissolved oxygen sensor based on fluorescence quenching of a ruthenium complex-doped sol-gel film was produced. Both planar substrate and optical fibre-based sensor configurations were shown to be viable. Phase fluorimetric DO sensing was also demonstrated. The sensor has excellent quenching characteristics ensuring high resolution at low oxygen concentrations, it possesses a short response time, and the stability and overall performance of the sensor is better than other optical DO sensors recently reported in the literature. The all-solid state sensor design indicates the potential for low-cost, high-performance, portable dissolved oxygen sensors for use in a wide range of measuring environments.

In the course of this work, a more fundamental understanding of the detailed quenching behaviour of the ruthenium complex by oxygen, both in gas and dissolved phase, was gained. Quenching behaviour was correlated with film microstructure and surface morphology, in terms of oxygen diffusion and solubility constants. The key materials development in this work was the identification of the optical sol-gel preparation parameters to yield high performance sensor films.

9.2 Future work

A prototype DO sensor based on the work reported in this thesis is being built at present. Meanwhile more extensive interferent studies need to be completed, and the long-term stability of the films in water needs to be monitored. These are important investigations to be undertaken prior to the commercialisation of the instrument. Optimisation of the barrier film, in particular the silicone rubber coating, must be pursued. The film must be thin enough such that it does not increase the response time significantly, while minimising fouling. Further investigations need to be carried out into the temperature dependence, and temperature compensation will be incorporated into the prototype.

Although a satisfactory sensor system has been constructed, the following fundamental issues remain to be addressed:

- The relative contributions of solubility and diffusion to the oxygen permeability,
- The effect of humid gas on films, with particular reference to gas-phase sensing,
- The relative contributions of purely aqueous phase quenching and gas phase quenching after partitioning,
- The temperature dependence of DO quenching,
- The separate contributions to response time and recovery time of the sensor
- The relationship between the hydrophobicity of ormosil films and the percentage of OH groups replaced by alkyl groups.

LIST OF PUBLICATIONS AND CONFERENCE PRESENTATIONS

Presentations:

Oral Presentations

- Development of a fibre-optic dissolved oxygen sensor based on quenching of a ruthenium complex entrapped in a porous sol-gel film. Conference on Chemical, Biochemical and Environmental Sensors VII, Munich, 19-23 June 1995.
- Development of an optical sol-gel-based dissolved oxygen sensor. Europt(r)ode III, Zurich, March 31-April 3 1996 [Invited paper - presented by supervisor].

Poster Presentations

- Tailoring of sol-gel films for optical sensing. Sensors and their Applications VII, IOP conference, Dublin, September 1995.
- Optimisation of sol-gel-derived silica films for optical oxygen sensing. 8th International Workshop on Glasses and Ceramics from Gels, Faro, Portugal, September 18-22 1995.
- Development of an optical sol-gel-based dissolved oxygen sensor. Sensors and Signals 3, Royal Society of Chemistry conference, Dublin, October 27-28 1996.

Publications:

Conference Publications

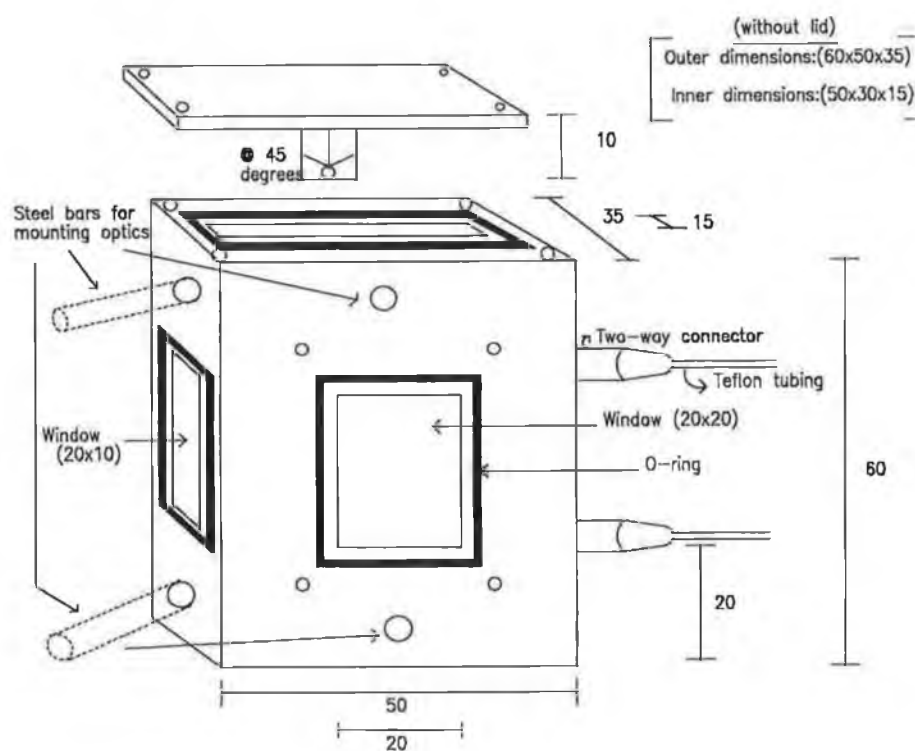
- Development of a LED-based fibre optic sensor using sol-gel-derived coatings. *Proc. SPIE*, 1994, Vol. 2293, pp. 110-120.
- Sol-gel coatings for optical chemical sensors. *Proc. SPIE*, Sol-Gel Optics III, San Diego, 1994, Vol. 2288, pp. 518-528.
- Development of a fibre-optic dissolved oxygen sensor based on quenching of a ruthenium complex entrapped in a porous sol-gel film. *Proc. SPIE*, Conference on Chemical, Biochemical and Environmental Sensors VII, Munich, 1995, Vol. 2508, pp. 190-198.
- Tailoring of sol-gel films for optical sensing. *Sensors and their Applications VII*, IOP conference, Dublin, 1995, Post-deadline paper.

Refereed Publications

- LED-based fibre-optic oxygen sensor using sol-gel coating. *Electronic Letters*, 1994, 30 (11), 888-889.
- LED-based oxygen sensing using evanescent wave excitation of a sol-gel immobilised dye. *Sensors and Actuators B*, 1995, 29, 1-3.
- Sol-gel coatings for optical chemical sensors and biosensors. *Sensors and Actuators B*, 1995, 29, 51-57.
- Light-emitting-diode-based oxygen sensing using evanescent wave excitation of a dye-doped sol-gel coating. *Optical Engineering*, 1994, 33 (12), 3861-3866.
- Dissolved oxygen sensor based on fluorescence quenching of oxygen-sensitive ruthenium complexes immobilised in sol-gel-derived porous silica coatings, *The Analyst*, 1996, 121, 785-788.

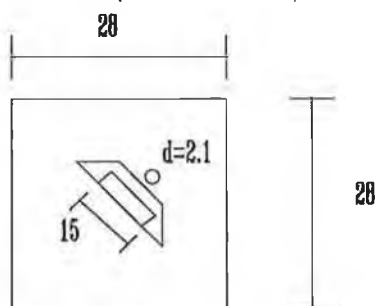
- Optimisation of sol-gel films for optical oxygen sensing. Accepted for publication in *Journal of Sol-Gel Science and Technology*, 1996.
- Optical chemical sensors based on sol-gel materials: Recent advances and critical issues. Accepted for publication in *Journal of Sol-Gel Science and Technology*, 1996.

APPENDIX 1(i): DESIGN OF INITIAL (23 mL) CHARACTERISATION CELL



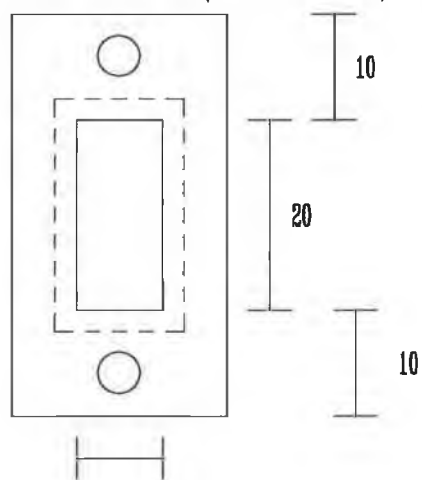
**APPENDIX 1(ii): DESIGN OF REDUCED-VOLUME (9 mL)
CHARACTERISATION CELL.**

TOP PIECE (OUTER DIMENSIONS)



Angle of wedge -- just
off 45 degrees

FRONT AND SIDE FACE (OUTER DIMENSIONS)



Inner dimensions
=15x15x40

APPENDIX 1(iii): DESIGN OF SMALL VOLUME (1 cm^3) GLASS CELL.

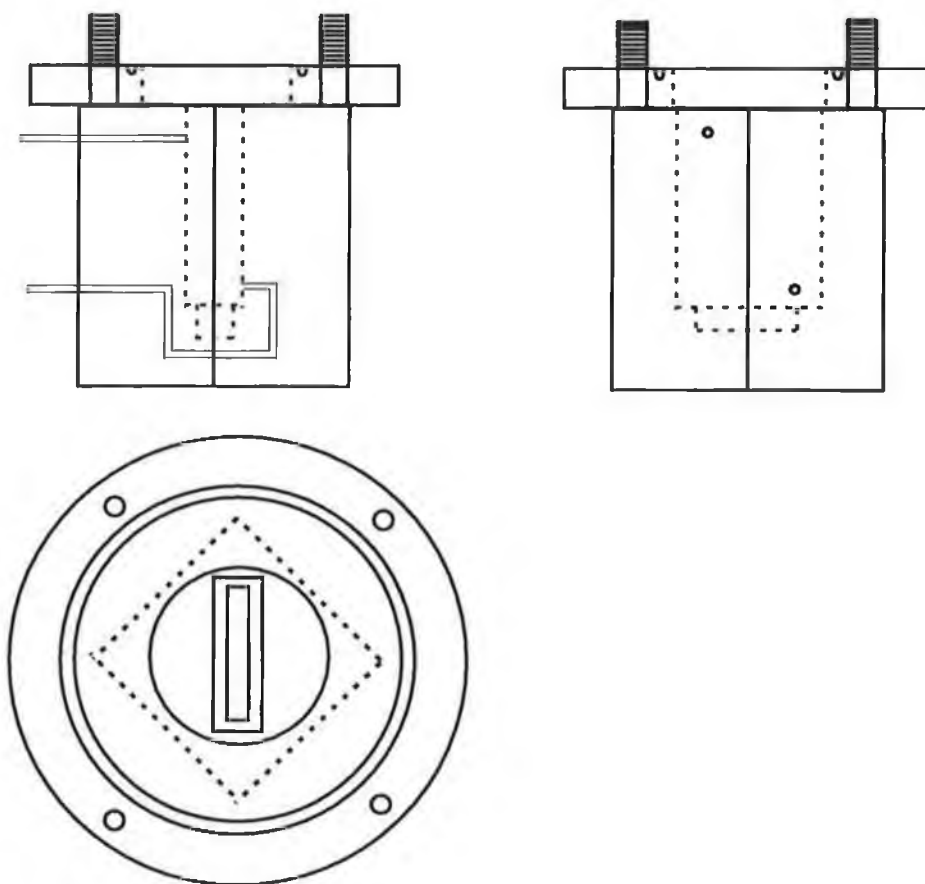


Figure (a.): Design for 1 mL volume glass cell

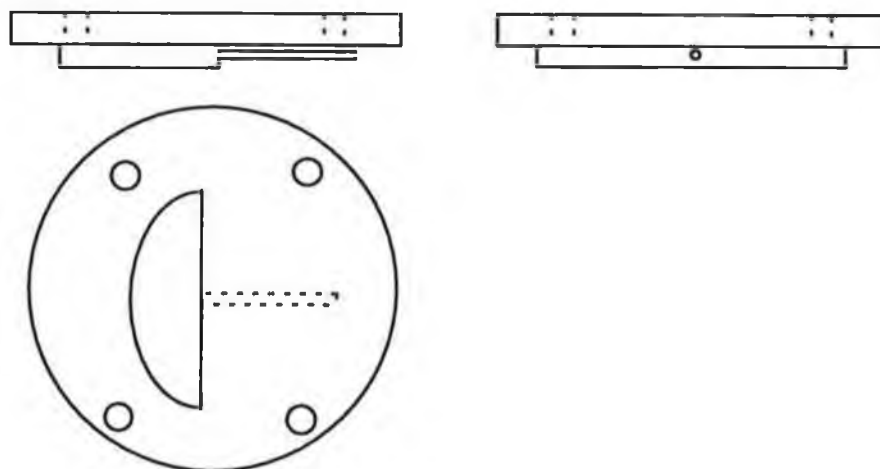
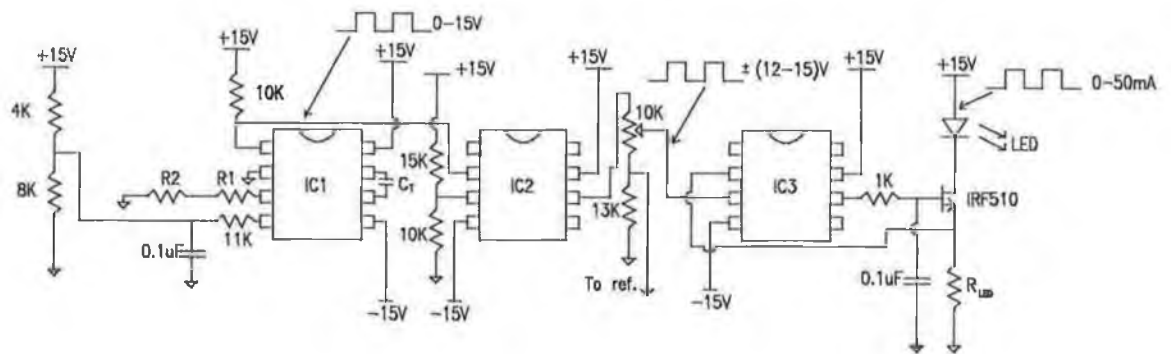


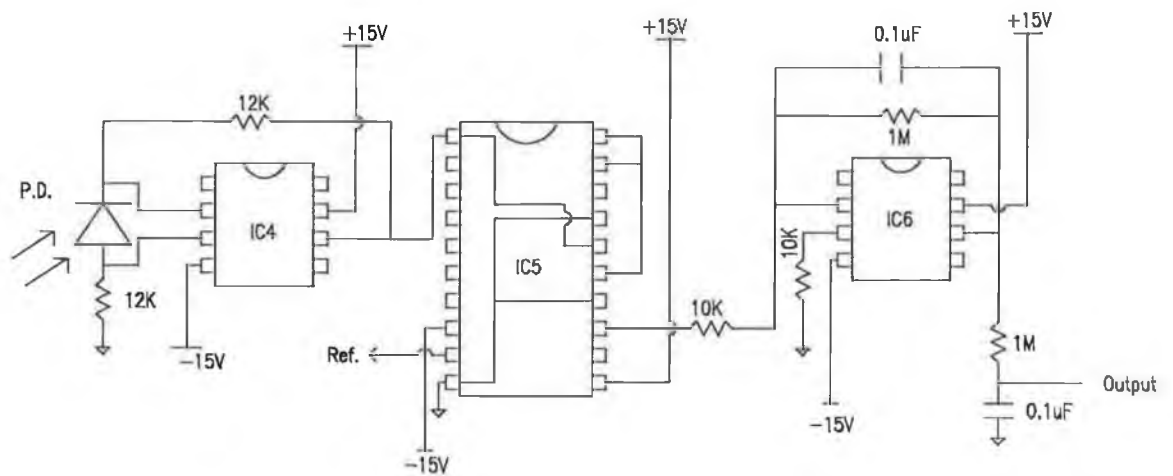
Figure (b.): Design for lid of 1 mL volume glass cell.

APPENDIX 2(i): LED DRIVING CIRCUITRY USING AD654



- IC1:** AD654, Voltage to frequency converter (RS#637-860)
IC2: LF351N, FET opamp (RS#308-528)
IC3: LF351N, FET opamp (RS#308-528)

APPENDIX 2(ii): AD630-BASED LOCK-IN AMPLIFICATION DETECTION CIRCUITRY.



IC4: CA3140E, BiMOS OpAmp (RS#308-130)

IC5: AD630, Balanced Modulator/Demodulator

IC6: As IC4